A COMPARATIVE STUDY OF LIPID PROFILE AMONG SMOKERS AND NON-SMOKERS

DISSERTATION SUBMITTED FOR

M.D. GENERAL MEDICINE

BRANCH – I

APRIL 2015

THE TAMILNADU
DR.M.G.R. MEDICAL UNIVERSITY
CHENNAI, TAMILNADU

CERTIFICATE

This is to certify that the dissertation entitled "**A COMPARATIVE STUDY OF LIPID PROFILE AMONG SMOKERS AND NON-SMOKERS**" is the bonafide work of **Dr. NASEEMA BANU.S.H**. in partial fulfillment of the university regulations of the Tamil Nadu Dr. M.G.R. Medical University, Chennai, for M.D General Medicine Branch I examination to be held in April 2015.

CAPT. DR. B. SANTHAKUMAR

M.Sc (F.Sc), M.D(F.M), PGDMLE, Dip.N.B (F.M),

The Dean,

Madurai Medical College,

Government Rajaji Hospital,

Madurai

CERTIFICATE

This is to certify that the dissertation entitled "**A COMPARATIVE STUDY OF LIPID PROFILE AMONG SMOKERS AND NON-SMOKERS**" is the bonafide work of **Dr. NASEEMA BANU.S.H** in partial fulfillment of the university regulations of the Tamil Nadu Dr. M.G.R. Medical University, Chennai, for M.D General Medicine Branch I examination to be held in April 2015.

Dr. S. VADIVEL MURUGAN M.D.,

Professor and Head,

Department of General Medicine,

Government Rajaji Hospital,

Madurai Medical College,

Madurai

Dr. C. DHARMARAJ M.D., D.C.H.,

Professor,

Department of General Medicine,

Government Rajaji Hospital,

Madurai Medical College,

Madurai

DECLARATION

I, **Dr. NASEEMA BANU.S.H.**, solemnly declare that, this dissertation "**A COMPARATIVE STUDY OF LIPID PROFILE AMONG SMOKERS AND NON-SMOKERS**" is a bonafide record of work done by me at the Department of General Medicine, Govt. Rajaji Hospital, Madurai, under the guidance of **Dr. C.DHARMARAJ M.D., D.C.H.,** Professor, Department of General Medicine, Madurai Medical college, Madurai.

This dissertation is submitted to The Tamil Nadu Dr. M. G. R. Medical University, Chennai in partial fulfillment of the rules and regulations for the award of M.D Degree General Medicine Branch-I examination to be held in April 2015.

Place: Madurai

Date: Dr. NASEEMA BANU S.H.

ACKNOWLEDGEMENT

I would like to thank **Capt. Dr. B.SANTHAKUMAR, M.Sc(F.Sc)., M.D(F.M)., PGDMLE., Dip.N.B.(F.M.).,** The Dean, Madurai Medical College, for permitting me to utilize the hospital facilities for this dissertation.

I also extend my sincere thanks to **Prof. Dr. S.VADIVEL MURUGAN M.D.,** Head of the Department and Professor of Medicine for his constant support during the study.

I would like to express my deep sense of gratitude and thanks to my Unit Chief and Professor of Medicine, **Dr. C.DHARMARAJ M.D., D.C.H.,** for his valuable suggestions and guidance throughout my study.

I express my sincere thanks to our beloved professors **Dr. V. T. PREMKUMAR, M.D., Dr. R. BALAJINATHAN, M.D., Dr. M. NATARAJAN, M.D., Dr. G. BAGHYALAKSHMI, M.D., Dr. J. SANGUMANI, M.D.,** and **Dr. R. PRABHAKARAN, M.D.,** for their interest in clinical teaching and constant support for me.

I express my sincere thanks to **Dr. S. GANESAN, M.D.,** Professor and HOD of Bio-chemistry for permitting me to utilize the facilities in the department for the purpose of this study and guiding me with enthusiasm throughout the study period.

I also offer my special thanks to the Assistant Professors of my Unit **Dr. P. S. Arul Raja Murugan, M.D., D.M.,** and **Dr. M. Rajkumar, M.D.,** for their help and constructive criticisms.

I thank **Dr. Akila, Dr. Ravi Kiran,** and **Dr. Sivakumar,** for their encouragement and help in my study.

I wish to acknowledge all those, including my other postgraduate colleagues, my parents and my husband who have directly or indirectly helped me to complete this work with great success.

Finally, I thank the patients who participated in the study for their extreme patience and co-operation without whom this project would have been a distant dream.

Above all, I thank The Lord Almighty for his kindness and benevolence.

ABBREVIATION

CAD	Coronary Artery Disease
ECG	Electrocardiogram
FFA	Free Fatty Acids
HDL	High Density Lipoprotein
IHD	Ischemic Heart Disease
LDL	Low Density Lipoprotein
MRFIT	Multiple Risk Factor Intervention Trial
TC	Total Cholesterol
TG	Triglycerides
VLDL	Very Low Density Lipoprotein
WHO	World Health Organization

LIST OF FIGURES

LIST OF FIGURES

LIST OF TABLES

CONTENTS

A COMPARATIVE STUDY OF LIPID PROFILE AMONG SMOKERS AND NON-SMOKERS

M.D. GENERAL MEDICINE

BRANCH – I

APRIL 2015

Dr. NASEEMA BANU.S.H.

ABSTRACT

Background & Objectives:

A prospective study was carried out to find the variations in lipid profile in smokers when compared to non-smokers. To study the alterations in lipid profile in terms of severity of smoking.

Materials and Methods:

This study was carried out among 200 patients who attended Government Rajaji Hospital, Madurai., for various ailments. The population was divided into 100 non-smokers and 100 smokers. The smokers were further divided into three groups depending on the intensity of smoking. Serum lipid profile was analyzed in all subjects.

Results:

Lipid Profile	Non-Smokers (Mean ± Std)	Smokers (Mean ± Std)	P-Value
TC	161.18±26.77	191.96±31.37	< 0.001
TG	103.58±26.26	164.29±28.95	< 0.001
HDL	49.58±8.57	44.72±9.96	= 0.002
LDL	82.34±16.57	103.08±18.66	< 0.001
VLDL	21.69±6.42	29.02±8.98	< 0.001

Dyslipidemia was directly proportional to intensity of smoking.

Interpretation and Conclusion:

Increase in total Cholesterol, Triglycerides, LDL & VLDL were found in smokers of all age groups. Where as HDL values showed inverse relationship. These changes were directly proportional to the severity of smoking. So, Tobacco smoking is associated with dyslipidemia which is atherogenic in nature.

Keyword:

Cigarette Smoking, Total Cholesterol, Triglycerides, Low density lipoprotein, High density Lipoprotein, Very low density Lipoprotein.

1. INTRODUCTION

Tobacco is one of the most potent and prevalent addictive, influencing behavior of human beings for over 4 centuries. Smoking is now rapidly increasing through the developing world and is one of the biggest threats to current and future health. Cigarette smoking is the most common type of tobacco use. Tobacco continues to be the second major cause of death in the world. Smoking is an important risk factor for atherosclerosis, peripheral vascular disease and stroke and cancers. It also has a strong relationship with gastric ulcer, periodontal disease and metabolic syndrome.

Cigarette smoking leads to the uptake of many hazardous compounds and their metabolites extracted from burning tobacco. The substances may be electrophilic and react with biological molecules giving rise to oxidative stress through the formation of reactive species or the initiation of lipid peroxidation chains in the membranes. Cigarette smoking has been found to alter the lipoprotein levels.

A one to three fold increase in risk of myocardial infarction has generally been noted among smokers. Plasma lipoprotein abnormalities are said to be the major underlying risk factor for the common occurrence of atherosclerotic vascular disease. Most of the studies indicate definite correlation between smoking and lipid profile alteration in which there is

definite dose response relationship between the number of cigarette smoking as well as the duration of smoking and changes in the lipid profile noted. However, in spite of all these information there is still much controversy about which part or parts in the lipid profile are mainly altered in response to cigarette smoking.

In the present study, an attempt has been made to find out the effect of smoking on the lipid in the health smokers are compared with that of same age group healthy non-smokers.

2. AIMS AND OBJECTIVES

- Comparative study of lipid profile among smokers and non-smokers.
- To study variable patterns of lipid profile in terms of intensity of smoking.

3. REVIEW OF LITERATURE

HISTORICAL ASPECTS

Tobacco has been used for centuries and possibly for millennia. At first it was smoked only by the native population of America and it was introduced to Columbus; but subsequently after tobacco was brought to Europe in the middle century, smoking became widespread throughout the world.

It was introduced to European society by Columbus after his maiden trip to the America in 1642. In Cuba he met a party of Tairo Indians with firebrand in their hands and herbs to inhale the smoke through the nostrils. This spread the use of tobacco from the American to Spain and European nation and ultimately all over the world.

When tobacco was first introduced into Europe, smoking was recommended for medicinal purposes; but its value soon became controversial. It was praised as prophylactic against many ills and condemned as a noxious vice, one of its notable opponents being James I, king of United Kingdom, who wrote vehemently against it and published in 1604 a treaty, a counterblast to tobacco, that smoking was harmful to brain, lungs, eyes and nose, however it was good for tax rolls.

Jeam Nicot extolled the medicinal values of tobacco which was supposed to cure various diseases including gout!. Thus tobacco became Nicotiana in honor of Nicot.

The association between atherosclerosis and tobacco smoking was noted by Buerger during earlier years of 20^{th} century. He observed ischemia of limbs (distally) among young smokers.

The risk of CAD decreases on cessation of smoking. This reduction in risk is significantly greater in a year of stopping smoking and there after gradually decreases until, after 10-20 years it is same as that of non-smokers. According to combined study of Alban and Framingham the smokers are at a 3 fold increased risk of acute coronary syndromes when compared with non-smokers of similar age groups.

In the past two decades, the rate of tobacco use was reduced in well-developed countries. So now the tobacco industries are targeting the nations, which are developing, and they are expanding their markets in those nations. About a third of the smoking population resides in china. This population is more than the smoking population in Europe and U.S. combined. According to WHO, by 2020, about three fourth of tobacco associated deaths will be occurring in the developing and under

developed nations. The death due to tobacco related illness crosses one million per year in china. This rate is 2.5 times more than that observed in U.S.

The effect on mortality is proportionally greater in male smokers than female smokers. But mortality in females is also increased when compared with non smoking females.

EPIDEMIOLOGY OF SMOKING

WHO has published a global fact-sheet regarding tobacco usage in countries as well as the effect of tobacco on mortality and morbidity. According to them

- Globally around one third of the adult males practice tobacco smoking.
- Smoking will be the cause of death in 1 among 6 persons by 2030 if the present trend persists. Globally 1 among 10 adults will be dying of smoking associated illness or it will be responsible for more than 4 millon deaths annually.
- One death occurs in each 8 seconds because of smoking tobacco.
- The habit of tobacco smoking is increasing greatly in developing countries. It tends to decline in well-developed nations. It was noted that there was a 50% reduction in the rate of smoking

tobacco among American population in the past few decades. The use of tobacco is rising by 3.5% every year in the developing countries.

- More than 9.8 million cigarettes are being sold each minute and more than 14.5 billion cigarettes are being sold every day.
- The tobacco related deaths in Britain citizens is 12 times more compared to the deaths from Second World War.
- 1 among 5 in American population die of tobacco associated illness.
- In WHO regions, maximum smoking rates were observed in west pacific areas. In these areas about 2/3 of adult males smoke.
- In the west pacific areas 1 among every 3 cigarettes are used.
- The tobacco trade is influenced mainly by the following corporations- japan, Britain and America.

YOUTH:

- Globally 1 adolescent in every 5 smoke.
- Everyday around 90,000 children begin smoking, globally. More than 50% of them are in Asian continent.
- It was observed that about half of those who begin smoking in teen age continue smoking for more than 20 years.

- Studies show that adolescents are much influenced by the advertisements for cigarettes.
- One fourth of the teenagers of west pacific areas will face death because of tobacco associated illness.

HEALTH

- Smoking 1 cigarette reduces five minutes in life span. About 50% of chronic smokers will be dying due to smoking associated illness.
- The single most preventable cause of premature death and illness is tobacco smoking. It happens to be the important factor in stroke, coronary artery disease and chronic obstructive pulmonary disease. It can cause cancers of the pharynx, larynx, oral cavity and lungs. It is also related to cancers of esophagus, stomach and urinary bladder. It is also related to cancers of pancreas, liver, ureters and kidney.
- Several harmful chemicals are found in the smoke from tobacco. A survey in Britain showed that more than 90% of females are unaware of the association of cancer of cervix with tobacco smoking.
- A survey was conducted in china, which showed that more than

sixty percent of adults in china were not aware of the association between cancer of lung and tobacco. About ninety five percent were not aware that smoking could cause coronary artery disease.

- About $1/4^{th}$ of deaths due to CAD and $3/4^{th}$ of deaths due to COPD are associated with smoking tobacco.
- More than one fifty million dollars are spent in U.S. for tobacco-associated diseases.

ADVERTISING

- US-based multinational Philip Morris-the world's biggest cigarette company-was the world's ninth largest advertiser in 1996, spending more than $3 billion.
- A survey a few years ago found that nearly 80% of American advertising executives from top agencies believed cigarette advertising does makes smoking more socially acceptable to children.
- Through advertising, tobacco firms try to link smoking with athletic prowess, sexual attractiveness, success, adult sophistication, adventure and self-fulfillment.
- A survey in the UK found half of smokers think that smoking "can't really be all that dangerous, or the Government wouldn't

have let cigarettes be advertised".

- A 1998 survey found that tobacco companies were among the top 10 advertisers in 18 out of 66 countries surveyed.
- In Asia, tobacco companies are among the top 10 advertisers Cambodia, Indonesia, Malaysia, Myanmar, and the Philippines.
- In Russia, according to press reports, foreign tobacco companies are the largest advertisers, accounting for as much as 40% of all TV and radio advertising.
- In 1997, the tobacco industry's spending on advertising in the United States was about $15 million a day ($5.7 billion for the year).
- The tobacco industry has changed the way it advertises in the last 30 years. Now only 10% of advertising expenditure goes to print, and outdoor advertisement, while more than half goes to promotional allowances and items, such as t-shirts for young people or lighters and key rings.

PGIMER Chandigarh recently carried out an analysis on the prevalence cigarette smoking in India. The study was carried out in 4 centers in India. The study group included people more than fifteen years of age. A detailed list of questions was prepared for the survey.

There were 11496(15.6%) ever smokers in the study sample of 73605 subjects. Among 37,682 males, 10,756(28.5%) were ever smokers and among 35,923 females, 740(2.1%) were ever smokers. Bidis were the most common form of smoking particularly in rural areas. The mean number of cigarette/bidis smoked per day was 14(±11.5) and the mean age of starting smoking 20.5(±20.0) years. The important agents associated with tobacco smoking were poverty and advancing age. The tobacco control programmes took strict measures against tobacco, which resulted in a cessation rate of around 11%. Around 15% of chronic smokers had respiratory problems. They concluded that a significant population in India has smoking habit (present or past) with the prevalence more in males compared to females. In spite of various anti tobacco measures the cessation rates were found to be very low. It was found that there was a significant smoking associated respiratory morbidity.

TYPES OF SMOKING

The practice of tobacco smoking is carried in a number of different ways to achieve one single goal: to obtain the nicotine kick, which is derived from the habit. Some of the different ways of smoking include:

- **Cigarette**
    - A paper wrapped cylinder, containing tobacco (which is

cured and finely cut) along with reconstituted tobacco and additives.

- May have filters at one end.

Fig.1. Cigarettes

- **Bidis**

Fig.2. Bidis

- These are thin, hand rolled cigarettes.

- Primarily produced in India and some Southeast Asian countries and imported to United States.
- Tendu or temburni leaf is used for wrapping the tobacco.
- A string is used for securing the ends.
- May be flavored or non flavored.
- Found to be more addictive when compared to normal cigarettes.
- More concentration of tar, carbon monoxide and nicotine are present.

- **Cigars, cigarillos and little cigars:**

Fig.3. Cigars

- Produced from air cured or dried tobacco.
- The tobacco leaves for cigar are initially aged for 1 year.
- Then in a multistep process, they are fermented.

- This process again can take upto 5 months.
- Bacterial and chemical reactions due to the process of fermentation produce alterations in the tobacco.
- This gives a distinct smell and taste to cigars.
- Compared to cigarettes regular cigars are larger. They wont be having filters.
- Little cigars or cigarillos will be similar to cigarettes. They will have filters and are filled with pipe tobacco. Sometimes they will be flavored.
- Compared to cigarettes, cigars have larger amounts of nicotine.

- **Kreteks:**

Fig.4. Kreteks

- These are often called clove cigarettes.

- Combination of cloves, tobacco, and other additives.
- Imported from Indonesia.
- The smoke contains large amounts of tar, carbon monoxide, and nicotine when compared to routine cigarettes.
- Users are found to have higher risk of acute lung injury.

- **Pipes**

Fig.5. Pipe

- Have a chamber, stem and mouth piece.
- Inside the bowl tobacco will be placed and lit.

- **Hookah:**
  - A type of pipe used for smoking shisha.
  - Tobacco along with vegetables or fruits is heated.
  - The smoke produced is filtrated through water.
  - The pipe has a head, body, water bowl and a hose.

- Charcoal is used for heating in the hookah.

Fig.6. Hookah

Electronic or E-cigarette:

Fig.7. E-Cigarette

- These are devices having a cadridge that is filled with nicotine, flavor and chemicals.
- The device is battery powered.
- This is a nicotine delivering system.

- This is not a tobacco product.
- The ingredients of e-cigarettes will be turned into vapour, which will be inhaled by the user.
- Here no tobacco burning is involved.
- No smoke will be produced.
- Only a fine heated mist is produced.
- Often resembles a real cigarette.
- Some other types look like a ballpoint pen.
- These are reusable.
- Replaceable and refillable cadridges are available.
- Available in different flavors and with different levels of nicotine.
- Marketed as an alternative and an aid in quitting tobacco.
- But no evidences are there to confirm the safety of e-cigarettes.

CONTENTS OF THE CIGARETTE SMOKE

1. **Gaseous substance**
    a. Hydrogen cyanide
    b. Carbon monoxide
    c. Nitrogen oxides

2. Volatile chemicals

a. Benzene

b. Acrolein

c. Formaldehyde

d. N-nitrosoamines

3. Sub-micron sized particle suspended in the smoke like

a. Phenol

b. Poly aromatic hydro carbons

c. Nicotine

d. Tobacco specific nitrosoamines

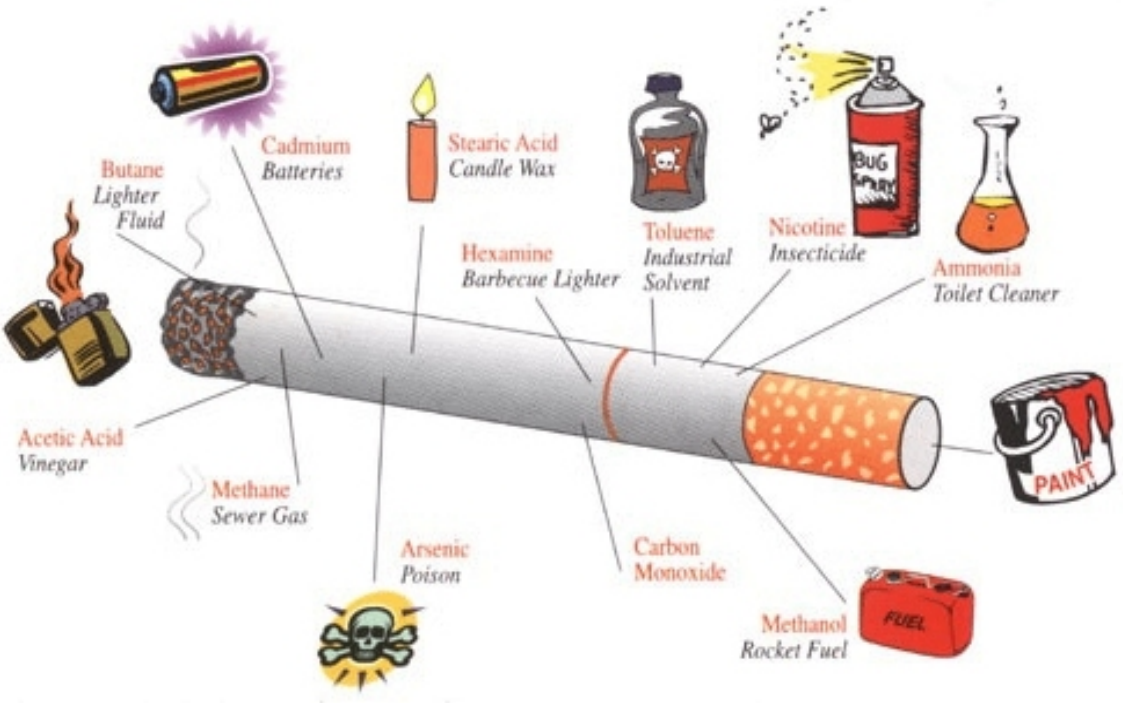


Fig.8. Contents of Cigarette Smoke

TABLE: 1. List of Contents in Cigarette Smoke

CHEMICAL	AMOUNT(PER CIGARETEE)
Acetaldehyde	980 micrograms to 1.37 milligrams
Acrylonitrile	Formally 1to 2 milligrams. This product was used as a fumigant in tobacco. Its use has since been discontinued
4-Aminobiphenyl	0.2 to 23 Nano grams per cigarette
O-Anisidine-Hydrocholoride	Unknown
Arsenic	Unkown
Benzene	5.9 to 75 micrograms
Beryllium	0.5 Nano grams
1,3-Butadiene	152 to 400 micrograms
Cadmium	1.7 micrograms
1,1-Dimethylhydrazine	Unknown
Ethylene oxide	Unknown
Formaldehyde	Unknown

Furan	Unknown
Heterocyclic amines	Unknown
Hydrazine	32 micrograms
Isoprene	3.1 milligrams
Lead	Unknown
2-Nahthylamine	1.5 to 35 Nano grams
Nitromethane	Unknown
N-Nitrosodi-n-Butylamine	3 Nano grams
N-Nitrosodiethanolamine	24 to 36 Nanograms
N-Nitrosodiethylamine	Up to 8.3 Nanograms
N-Nitrosodimethylamine	5.7 to 43 nanograms
N-Nitrosodi-n-Propylamine	1 Nanogram
4-(N-Nitrosom-ethylamine)-1-(3-Pyridyl)-10Butanone	Upto 4.2 micrograms
N-Nitrosonomicotine	14 microgram

N-Nitrosopiperidine	Unknown
N-Nitrosopyrrolidine	113 Nanograms
N-Nitrososarcosine	22 to 460 nanograms
Polonium-210	Variable, depending on soil and fertilizer used to grow tobacco
Polycyclic hydrocarbon	28 to 100 milligrams
O-Toluidine	32 Nanograms
Vinyl chloride	5.6 to 27 Nanograms

CHARACTERISTICS OF SMOKERS

Many features distinguish smokers from non-smokers

- They tend to have increased amount of tea, coffee
- They consume more alcohol
- They have reduced exercise performance
- They have weaker immune mechanisms
- They have a small increase in the total white blood cell count, hematocrit, serum IgE levels, and platelets count.

- They have reduced levels of leucocyte ascorbic acid.
- Their serum levels of albumin are less compared to non smokers.
- Their serum uric acid levels are less
- They have reduced HDL to LDL ratio
- Their leucocyte counts show a strong inverse relationship with HDL cholesterol, a positive association with triglycerides and LDL cholesterol in smokers.
- There is reduced synthesis of PGI2, a potent vasodilator, a cyclooxygenase product of vascular endothelium and an inhibitor of platelet aggregation.
- They have increased number of pulmonary macrophages which show altered metabolism
- They are found to have abnormal results in lung function tests.
- Tobacco smoking is found to stimulate hepatic microsomal enzyme systems and interact with a lot of drugs like
    - Propranolol-augmented first pass clearance
    - Propoxyphene-reduced analgesic effect

- Benzodiazepines-reduced sedative effect
- Beta blockers-heart rate and blood pressure lowering effect are reduced
- Chlorpromazine-reduced serum levels
- Oral oestrogens-increased clearance by liver
- Haloperidol-reduced serum concentrations
- Heparin-faster clearance
- Insulin- reduced absorption due to cutaneous vasoconstriction.

SMOKING AND LIPID PROFILE

The various mechanisms postulated in the alteration of lipid profile among smokers are

- Stimulation of sympatho adrenal system by nicotine leading to lipolysis and increased serum free fatty acid(FFA) levels which leads to increased synthesis of VLDL from the liver , hence increased triglycerides(increased consumption of FFA by heart that leads to increased myocardial oxygen demand).
- HDL concentration varies inversely with VLDL concentration in serum.

- Repressive action of smoking on oestrogen level in turn leads to decreased HDL cholesterol

- Smokers are thought to consume diet rich in fat and cholesterol and poorer in fibres and serials.

- Difference in oral fat handling between smokers and non smokers.

Smoking of tobacco by people, started centuries ago but the health and environmental hazards posed by it was recognized only in the 20th century. Atherogenesis leading to coronary artery disease and cerebrovascular accidents is thought to be accelerated by smoking. The exact atherogenic mechanism of smoking is still unclear. It has been observed by workers that smoking leads to dyslipidemia which is a major factor for atherosclerosis.

Gofman JW et al were the early pioneers of this study and had found that cigarette smoking and serum lipoproteins had a strong association between themselves as well as coronary artery disease. The initial examination revealed that 82 subjects had clinically evident CAD. They were excluded from the sample leaving a total of 5127. 4469 (69%) of the 6567 in the initial sample actually underwent the first examination. After the first examination, the study population was examined at intervals of two

years. Information was obtained with regard to serum cholesterol, blood pressure, weight and cigarette smoking. Although biennial examination was the main source of follow up information, other means were also adopted to detect CHD (example: death certificate records).

Among other things, the study showed that increasing serum cholesterol values are associated with increased CHD risk. The study also showed that the association between smoking and CHD varied with manifestations of the disease. Thus smoking was most strongly associated with sudden death from CAD than with less fatal forms of the disease. Risk factors have been found include male sex, advancing age, high serum lipid concentration, high blood pressure, cigarette smoking, diabetes mellitus, obesity, low vital capacity and certain ECG abnormalities. The predictive value of serum lipids, blood pressure and cigarette smoking were repeatedly demonstrated. The Framingham heart study became a prototype of similar studies in US and other countries.

FELIC study showed that development of coronary artery disease (atherosclerosis) begins in childhood itself. It is therefore important to identify potential risk factors early when prophylactic care can be cost effective (example; cigarette smoking).

Wendy H Graingetal demonstrated significantly higher serum levels of triglycerides(+11.8%), VLDL(+12.4%) and LDL(+4.1%) and lower level of HDL(-8.5%) and TC(-3-7%). This study was in the age group of 8-19 years old. These changes were comparable with the changes seen in adult smokers except for total cholesterol, which is usually high in adult smokers. Misawa K and his colleagues ascertained the relationship among HDL, cholesterol and other serum lipids with active and passive smoking, obesity, alcohol drinking and working status in healthy adults and in school children sampled at random who were medically healthy. They found that triglycerides were high and had a direct relation to smoking while high-density lipoprotein were low and had an inverse relationship to the number of cigarettes.

Freedman DS et al from Bogalusa Heart study reported that in smokers following a fat rich meal, concentration of triglycerides increases to a greater degree and remains elevated for longer duration. Smoking promotes atherosclerosis by several mechanisms. Richmond W et al from st.mary's hospital, London proposed that HDL changes and reduced removal of TGL rich lipoproteins, expose the endothelium of blood vessels to the atherogenic lipoproteins and there is increased predisposition to develop CHD.

Jensen J and Christiansen C in their study on serum lipoproteins during hormone replacement therapy and effects of smoking, found that total serum cholesterol was significantly reduced in smokers and non smokers but the response in smokers was only half of that observed in non smokers. Difference was also noticed in subjects with different modes of hormone administration. Response was better in those using the percutaneous route than the oral route.

Largue G etal reported one experimental study with the use of nicotine gum on variation in total cholesterol and LDL cholesterol. They studied lipid modification in 14 voluntary students, heavy smokers (more than 20 cigarette per day) and nicotine dependents (Fager storm test scores > 7). They received in double blind study, either 2mg nicotine gums or placebo gums (8-12 gums a day for one month). Total cholesterol, HDL cholesterol, LDL cholesterol and triglycerides were measured at day 0 and day 60. They found that at day 60 HDL and LDL cholesterol were significantly improved in those who gave up smoking. It was noted that there was also improvement in nicotine dependent smokers who were on nicotine gums.

Gerogesteiner and his associates, who studied the pathophysiology and natural history of atherosclerosis of coronary artery and its association

with elevated level of intermediate density lipoproteins, found that triglycerides were high among the groups with increased rate of cigarette smoking.

Moffatt RJ from the department of nutrition, food and movement science, Florida reported on increase of HDL cholesterol levels after cessation of smoking. Tilwani RK and his associates who studied total cholesterol, triglycerides, LDL, VLDL, and HDL found that TGL, LDL, VLDL and cholesterol values were elevated among smokers than non smokers.

Rastogi R and his associates studied lipid profile in 51 healthy male non-smokers and 152 healthy male smokers. In this study they included both beedi and cigarette smokers. Mean value of total cholesterol LDL, VLDL, triglycerides were high in all groups of smokers but the rise was more in heavy smokers and in persons who smoked for more than 20 years. When compared with non-smokers the mean value of serum HDL and cholesterol was lower in all groups of smokers. The difference was maximum in heavy smokers.

Tiwari AK and his associates studied changes in body lipids in healthy people and CHD patients. 51 normal volunteers and 34 clinically established coronary heart disease patients were studied. 21 out of the 51

normal and 16 out of the 34 coronary heart disease patients were cigarette smokers. The cases were divided into two groups, Subjects aged 20-40yrs, the younger age group and 41-61 yrs., the older age group. TC and HDL cholesterol of all cases were determined .the ratio of total cholesterol to HDL cholesterol was significantly higher in all the normal and coronary heart disease smokers. Hence the higher level of ratio of total cholesterol to HDL an important parameter helps to ascertain the development of coronary heart disease in cigarette smokers.

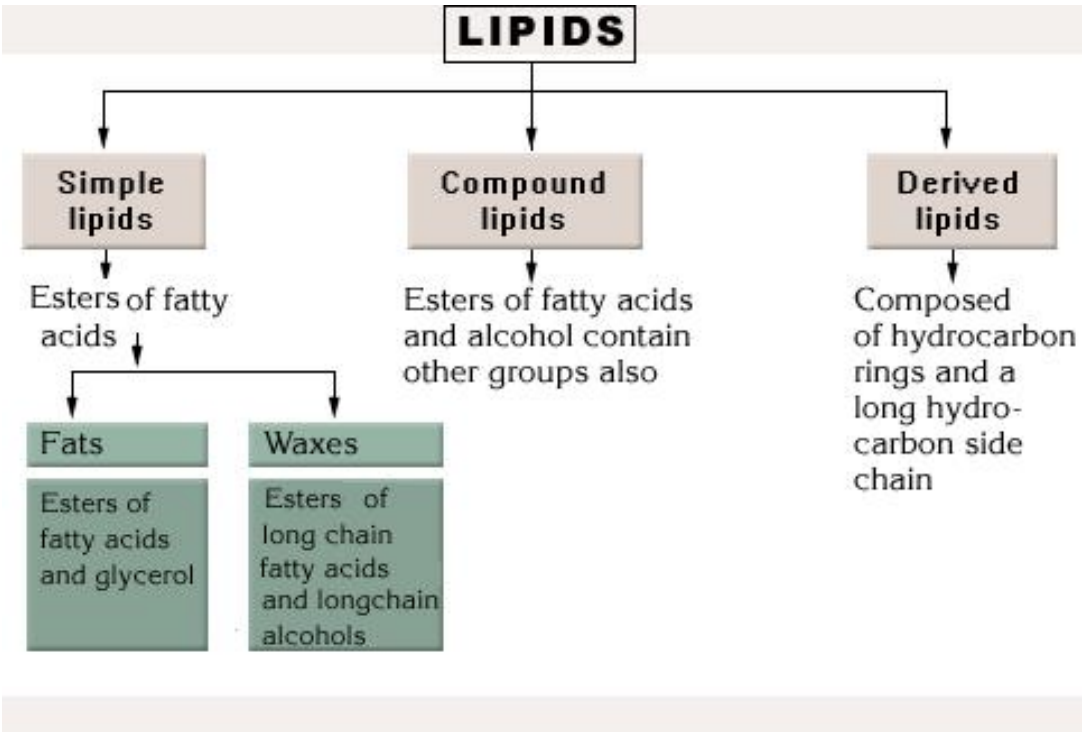
A study by Khurana M and associates on lipid profiles in cigarette smokers and tobacco chewers showed HDL cholesterol to be lower in these people when compared to controls. It was also found that both tobacco chewers and cigarette smokers have an equal dyslipidemia and there is a raised risk for coronary artery disease.

PLASMA LIPIDS AND LIPOPROTEINS

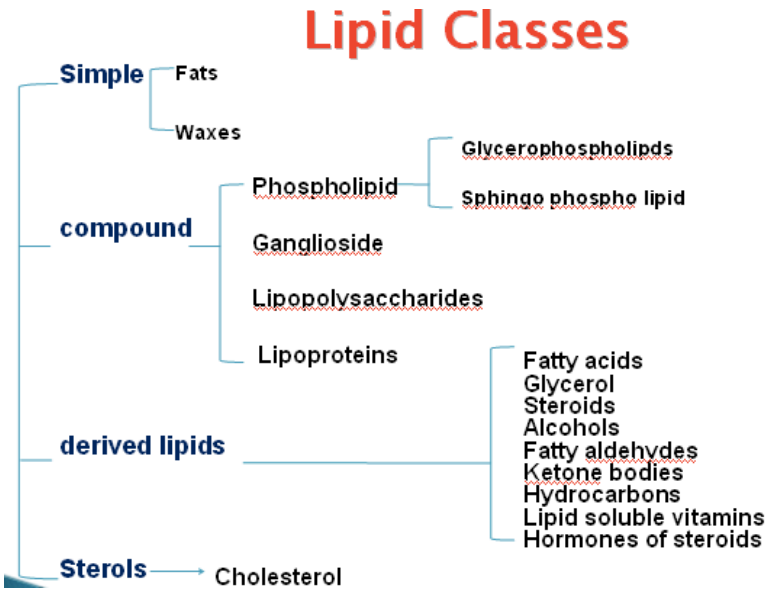
Chemistry of lipids:

The lipids constitute a wide range of substances of biological origin. These are soluble in organic solvents and are poorly soluble in water. Metabolically lipids are more closely interrelated, being transported together in the plasma lipoproteins and sharing certain mechanisms

Lipids can be classified as follows:



(a)



(b)

Fig. 9. (a) & (b) Classification of Lipids

Fatty Acids

They are present as such in minute concentration in plasma and cells. They are constituents of most lipid classes. They comprise a carboxylic acid group and a chain of hydrocarbon and are thus aliphatic monocarboxylic acids. The fatty acids are arbitrarily divided into those with long chain, medium and short chains. They are also grouped into saturated and unsaturated fatty acids depending on the absence or presence of a double bond respectively. The main saturated fatty acids are palmitic and stearic acids. Most of the fatty acids are carried mainly by albumin. Essential fatty acids are those, which cannot be synthesized in the body. Free fatty acids are immediately available energy sources and provide much of the energy requirements of the body. (Normal values range from 250-400 mg/dI)

CHOLESTROL:

Cholesterol is a word from Greek. Chole=bile, sterols=solid, ol=alcohol. It is synthesized in all nucleated cells. 120 gram of cholesterol is available in a 60 kilogram man

- Nervous system contains 30 gm
- Muscles contain 30 gm
- Adipose tissue has 30 gm
- Skin contains 30 gm

- Blood has 10 gm
- Spleen and liver contain 10 gm
- Bone morrow has 5 gm
- Alimentary tract has 3 gm
- Adrenal gland has 2 gm

It is a crystalline solid with yellow color. They have notched appearance when examined under microscope. It is soluble in chloroform and other fat solvents. It is absent in plants and prokaryotes, present in all animal tissues. It was associated from bile stones by Poulletier de la sale in 1758. Chevreul ME characterized it in 1818. Heinrich Wiela got Nobel price in 1927 for enunciating the complete structure of cholesterol in 1918.

FUNCTIONS:

- Important component of cell membrane
- Important in nerve conduction by its insulating effect on nerve fibres.
- Important precursor for bile acids and salts.
- Androgens, estrogens and glucocorticoids are derived from it.
- 7-Dehydrocholesterol is the precursor for the synthesis of vitamin D3.
- Cholesterol esters are formed when OH group of cholesterol is esterified to fatty acids. This occurs in the body by transfer of PUFA moiety by lecithin acyl transferase.

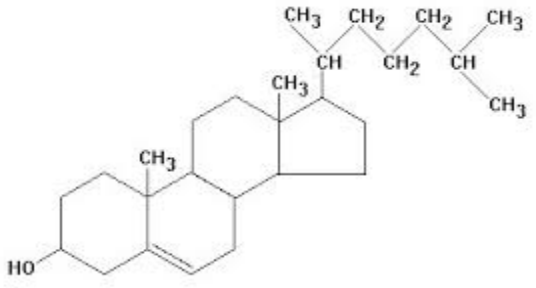


Fig. 10. Structure of Cholesterol

BIOSYNTHESIS OF CHOLESTEROL

All the carbon atoms for cholesterol synthesis are acquired from acetyl COA. The pathway for synthesis was described by sir John Comfort and Vladimir Prelog. In 1975 they got Nobel price for this. It is synthesized in many parts of the body like liver, adrenal cortex, ovaries, testes, and intestine. Nucleated cells including walls of arteries can synthesize cholesterol. The various enzymes required for the synthesis of cholesterol are available partly in cytoplasm and partly in endoplasmic reticulum.

EXCRETION OF CHOLESTEROL

300 mg of cholesterol is present in an average diet. 700 mg of cholesterol is synthesized in the body in one day. 500 mg of cholesterol in excreted in bile and part of this is reabsorbed in intestines. Intestinal bacteria act upon the un absorbed cholesterol to form cholesterol and coprostanol(fecal sterols). The remaining 500 mg is used in forming bile acids.

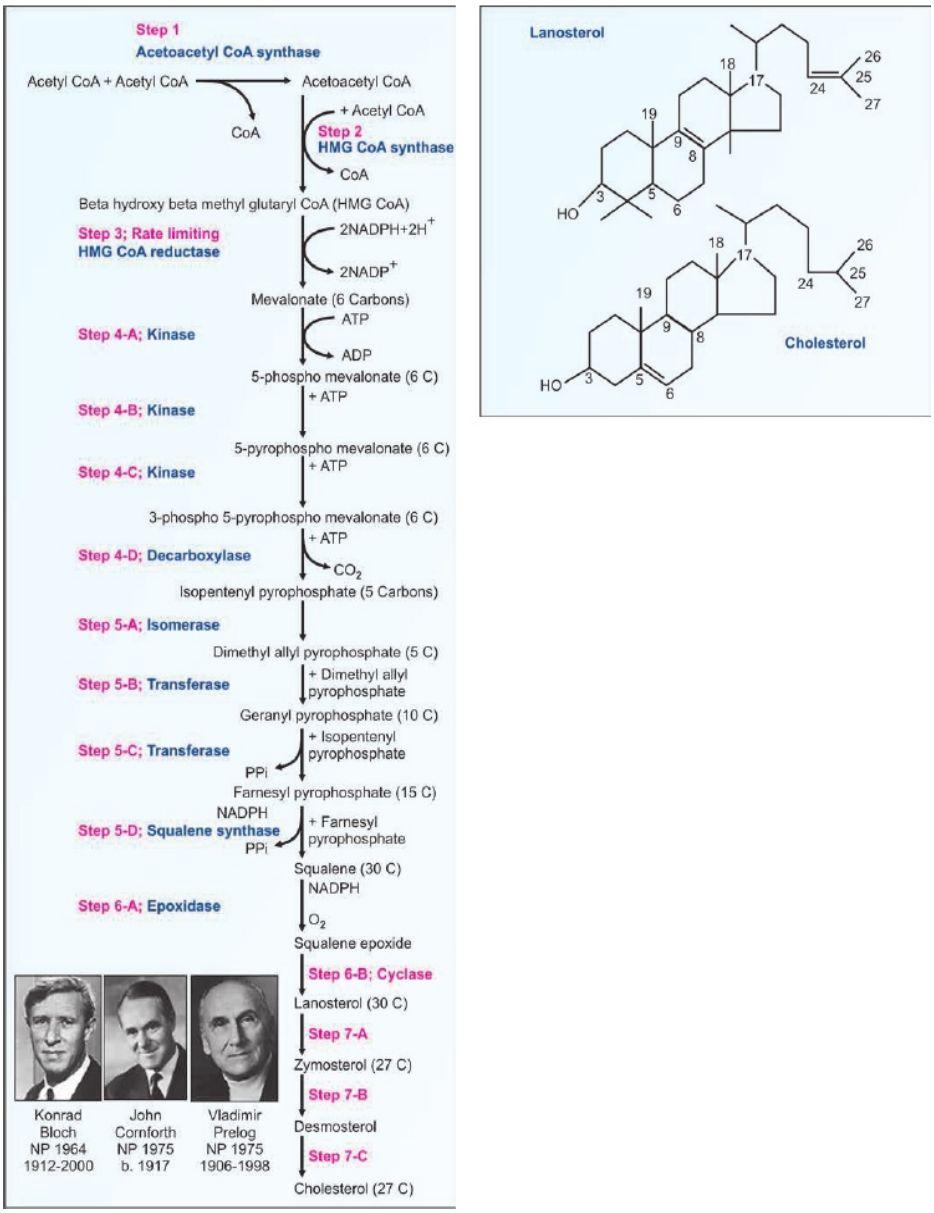


Fig. 11. Biosynthesis of Cholesterol

CHOLESTEROL AND LIVER:

Liver can synthesize cholesterol. It removes cholesterol from lipoprotein remnants. It forms bile acids from cholesterol. It excretes cholesterol through bile.

TRIGLYCERIDES

These are compounds of one molecule of glycerol united by ester bonds to fatty acids (3 molecules). If any 1 or 2 among those 3 hydroxyl groups of glycerol is esterified then they are called mono or triglycerides respectively. Triglycerides are an important energy store. Plasma triglycerides are derived from two sources. Exogenous, or dietary triglycerides are derived from food and circulate in the plasma in the form of chylomicrons. These are large, low-density particles, which are formed in intestinal epithelial cells and appear in the plasma soon after a meal. Chylomicrons deliver dietary triglycerides to the adipose and other tissues. In normal individuals, they are entirely cleared from plasma within 12 hours of fasting.

Endogenous triglycerides are derived primarily from hepatic conversation of carbohydrate and amino acids. Eighty percent of it circulates in plasma in the form of very low-density lipoproteins (VLDL), which transport triglycerides to adipose and other tissues. As triglycerides are removed, intermediate density lipoprotein is formed, some of which undergoes catabolism in the liver to low-density lipoprotein (LDL) particles. A small proportion (approximately 15 percent) of plasma triglycerides are carried in LDL and a tint fraction is contained in the high-density lipoprotein

(HDL) moieties. 1984 consensus panel divided plasma triglycerides levels into three categories: “normal” was defined as a TG level less than 250 mg/dl and HTG was dichotomized into “borderline hypertriglyceridemia” (250 to 500 mg/dl) and “true hypertriglyceridemia” (>500mg/dl). Levels of TG greater than 100 mg/dl are usually due to chylomicronemia and are believed to be able to cause recurrent abdominal pain and pancreatitis.

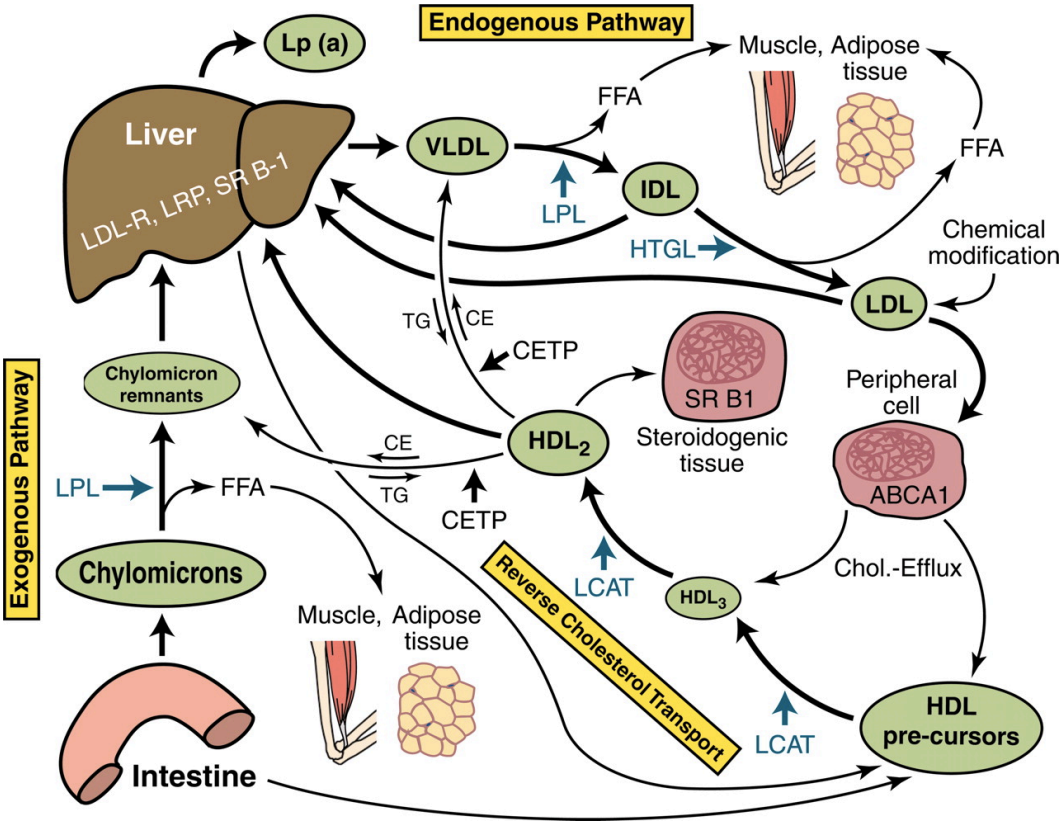


Fig.12. Lipid Metabolism

Elevated triglycerides are clinically important owing to their direct relationship to pancreatitis, and their association with glucose intolerance and renal and hepatic disease. Triglycerides measurements, as part of lipid

evaluation for atherosclerotic risk, are important as they provide the only convenient and cost-effective method for routinely estimating LDL cholesterol. Their inverse relationship to HDL makes triglyceride measurements a cost effective screening procedure. Unlike cholesterol measurement, triglycerides should always be assessed in the fasting state.

PHOSPHLIPIDS

They are complex lipids resembling triglycerides, but in addition to glycerol and fatty acids, they contain one or more phosphoric acid groups and a nitrogenous base. The major phospholipids in plasma are lecithin and sphingomyelin. The phosphate and nitrogenous base are water soluble, a fact that is important in lipid transport. (Normal values range between 150-275 mg%).

LIPOPROTEINS OF PLASMA

Lipids are insoluble in water. When lipids combine with water-soluble complex proteins, they become soluble and constitute lipoproteins. The role of protein-lipid complexes in maintaining the lipids in solution in plasma was suspected by Schulz(1897) and Nerking (1901) at the turn of 20th century. Application of new physical methods for protein separation, including electrophoresis and ultracentrifugation expedited the progress in lipoprotein chemistry. Tiselius et al in 1941 reported the existence of two

lipoprotein classes, separable by moving boundary electrophoresis. These were alpha and beta lipoproteins. It was another decade before a further component; Pre-B lipoprotein was identified by zonal electrophoresis (DangerfieldWO,1955).

CLASSIFICATION:

- Chylomicrons contain apolipoprotein B-48.
- Very low density lipoproteins or pre beta lipoproteins. B-100 is the important apoprotein.
- Intermediate density or broad beta lipoproteins.
- Low density or beta lipoproteins. Major apoprotein is B-100.
- High density or alpha lipoprotein. Major apoprotein is apo-A.

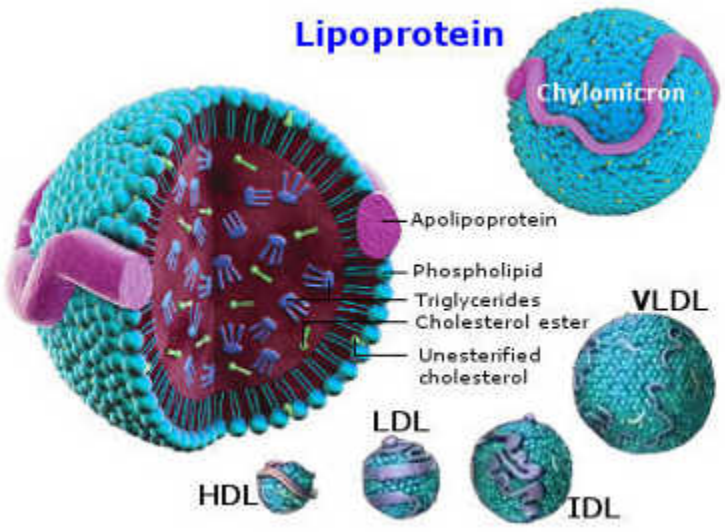


Fig. 13. Lipoprotein Structure

Among the many technique used to separate lipoprotein, the following are important

1. **Ultracentrifugation**

There are two principle ways of using ultracentrifugation to determine lipoproteins. They employ two different instruments. The preparative and the analytical centrifuge.

a) **Preparative Ultracentrifugation**: Plasma has a salt density of about 1.006. Ultracentrifugation of plasma without adjustment of its density for a short period brings the chylomicrons rapidly to the top of the tube. Longer ultracentrifugation at this density allows the VLDL to be collected on the surface. Addition of salt to plasma will further raise the density to selected levels and permit isolation of other lipoproteins. Those lipoproteins, which are separated below a density of 1.006 constitute VLDL, 1.006 to 1.063 constitute LDL, 1.063 to 1.21 constitute HDL.

b) Analytical Centrifugation: In this instrument, plasma fraction usually prepared at salt density of 1.063, is centrifuged at high speeds and the moving bands of the floating lipoproteins are photographed and later used to determine the concentration that are referred to certain standard conditions.

2. **Electrophoresis:**

If the plasma is examined by paper or agarose gel electrophoresis at pH 8.6, it is possible to demonstrate, by means of fat stains, the existence of four bands. Three of these move towards the anode and one remains at the origin, that is, the line of application of the serum, the faster moving band occurs approximately in the same position as alpha-I globulin. It is known as alpha-lipoprotein and corresponds to the HDL fraction, demonstrated by ultracentrifugation floatation technique. The next band appears at approximately the position of beta globulin and is known as beta lipoprotein, which corresponds to LDL fraction. Another band appears between the position of alpha globulin and beta globulin. It is known as pre-beta lipoprotein, which corresponds to VLDL fraction. The band, which remains stationary at the origin, consists of the chylomicrons. The five principal lipoprotein classes are defined according to their density on ultracentrifugation and by their mobility on agarose gel electrophoresis. In addition, they can be classified on the basis of size and relative concentration of cholesterol or triglyceride and by their apoprotein content.

Chylomicrons

Chylomicrons are the largest of the lipoproteins. Their primary function is to transport dietary, or exogenous, triglycerides and cholesterol

from the intestinal lumen to the sites of metabolism or storage. The chylomicrons are formed in the gastrointestinal (GI) tract. In the lumen of the GI tract, dietary fat is degraded into free fatty acids and monoglycerides. These substances enter the intestinal villi, where they are reconstructed into a triglyceride particle. Dietary cholesterol absorbed into the intestinal wall is then esterified to cholesteryl esters, mainly cholesteryl oleate, by the enzymatic reaction catalyzed by cholesterol acyl transferase. The triglyceride and cholesteryl esters are then combined with apos beta-48, A-I and A-IV within the intestinal wall to form chylomicron particles.

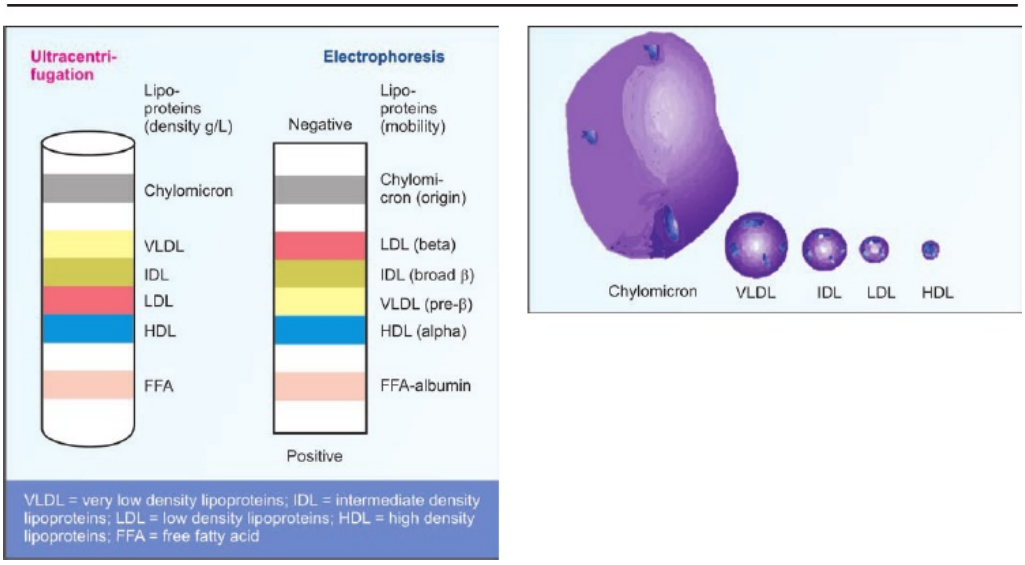


Fig. 14. Lipoprotein Separation Techniques

The nascent chylomicrons enter the systemic circulation by the way of lymphatics. Apos E and C are then added to the particles. Normally, the chylomicrons are cleared rapidly from the blood and are virtually absent in

the fasting state. The clearing of the chylomicrons is modulated by the enzyme lipoprotein lipase (LPL). LPL catalyzes hydrolysis of the triglyceride core of the chylomicron, leaving a remnant particle rich in cholesterol, apo C, apo E, and apo B-48. During this process, apoproteins, phospholipids, and cholesterol from the surface of the chylomicron are transferred to HDL particles. The chylomicron remnants are cleared rapidly from the circulation by receptors present on the surface of liver cells. These receptors recognize the apo E component of the remnant particles. Remnants that contain the apo E2moiety bind less well, and are thus removed less quickly than remnants that contain either the apo E3 or E4 moiety.

Chylomicron remnants are thought to be atherogenic, and an abnormal delay in their clearance is therefore undesirable. Delay in chylomicron clearance may be secondary to a genetically inherited deficiency of LPL or its activator, apo C-II. It is partial degradation of the chylomicron to a remnant that renders the particle atherogenic. Delayed clearance of the remnant particles may damage the vascular endothelium, and thus predispose to atherosclerosis.

Hyperchylomicronemia also may be secondary to other acquired hypertriglyceridemic states, such as those seen with exogenous estrogen

use, uncontrolled diabetes, and excessive alcohol intake. The presence of chylomicrons in the serum is necessary for the diagnosis of typeI or V hyperlipoproteinemia in the Fredricksons classification system.

Very Low-Density Lipoprotein

VLDLS are the intermediate in size between chylomicrons and IDLS. They are relatively large particles, with diameters ranging from 500 to 800 A. VLDLS are produced in the liver. Their primary lipid component is triglyceride, but cholesterol, cholesterol ester, and phospholipid are also present. The synthesis of VLDL is increased by excess carbohydrate, alcohol, or caloric consumption. VLDL is used to take the cholesterol & TGL to the pen tissues & have fatty acids are utilized for energy & also stored as TGL.

When VLDL particles enter the systemic circulation, their triglyceride core is hydrolyzed by LPL. As the VLDL particle is degraded, most of its surface apoproteins, except for apo B-100, are transferred with other surface components to HDL. The remaning VLDL remnant is called IDL. Unlike the chylomicron remnant, IDL contains apo B-100 rather than pro B-48. The metabolism of VLDL is complex and not fully understood. Some of the larger particles appear to be directly removed from the circulation. The rest

of the particles enter the cascade, in which they are converted to IDL and eventually to LDL.

INTERMEDIATE DENSITY LIPOPROTEIN

IDLs, which carry both cholesterol and triglyceride, are the products of the enzymatic (LPL-medicated) breakdown of VLDL. After their formation, IDLs may be removed by the liver by means of the binding of apo E to the IDL or B/E receptor. The remainders are converted to IDL, a process, thought to be mediated by hepatic triglyceride lipase. IDLs have high cholesterol content and migrate in the beta region on electrophoresis. Elevation of IDLs is thought to predispose to premature CAD and peripheral artery disease. Accumulation of IDL is characteristic of dysbeta lipoproteinemia, also called Fredrickson's type III hyperlipoproteinemia. This relatively uncommon from hyperlipoproteinemia is associated with both triglyceride and cholesterol elevation.

LOW DENSITY LIPOPROTEIN

LDL, which is 45 percent cholesterol by weight, is the major carrier of cholesterol to the nerve tissue, cell membranes and other cells that require the cholesterol for metabolic functions, including the synthesis of steroid hormones. LDLs have a density of 1.019 to 1.063 gm.ml a diameter of 180 to 280 A, and beta electrophoretic mobility. LDL usually is formed from

VLDL breakdown. Direct synthesis has not been completely excluded. Increase LDL synthesis may occur be means of enhanced conversion of VLDL remnants or direct hepatic production of apo B containing lipoproteins.

ApoB-I00 is the only protein found in LDL, and makes up about 20 percent of the LDL mass. Each particle is thought to contain 1 molecule of apo B- 100, but the ratio of protein mass to total particle mass can vary from the large to the small particle range. LDL particles are heterogenous, differing in their hydrated density and cholesteryl ester in the LDL particle, for example, may vary upto 40 percent by weight. Patients with greater concentration of small, dense LDL, have been reported to have a three times greater risk for acute myocardial infarction (MI), regardless of weight or gender. Small, dense LDL molecules are commonly associated with male gender, diabetes, depressed HDL levels and familial combined hyperlipoproteinemia.

LDL particles are recognized by specific LDL or apo B/E receptors on the surfaces of hepatic and certain non hepatic cells. These receptors also recognize and bind some of the apo E containing IDL particles, preventing their conversion into LDL. Bound LDL particles (and IDL particles) are then internalized into the cells. About 75 percent of the LDLs in the blood stream

are removed by this specific receptor mediated binding. The reminding LDL particles are cleared by scavenger or macrophage receptors or by non-receptor mediated mechanism. The number of LDL receptors is not fixed, and can be modified by genetic defects, saturated fat and cholesterol intake or certain pharmacological agents.

The prototype disease involving the LDL receptor is familial hypercholesterolemia. In this condition, heterozygotes have a 50 percent reduction in LDL receptors, where as homozygotes have little or no receptor activity. Familial hypercholesterolemia is fairly common, occurring in 1 in every 500 people. Familial combined hyperlipidemia (FCH) is even more common, possibly occurring in 1 in every 500 people. Clinically, FCH patients may be difficult to differentiate from those with familial hypercholesterolemia. In FCH, most patients lack tendon xanthomas; most family studies show varying Fredrickson's phenotypes.

The characteristic defect in FCH is thought to be an overproduction of apo B- 100 by the liver. Also, FCH patients have a lower ratio of apoA-I to apoB-100. About 80 percent of patients with an elevated LDL value do not have only one gene defect; the dyslipidemia is secondary to polygenic factors. Hence, elevations of LDL due to primary receptor defects are relatively uncommon.

Fredrickson Classification of the Hyperlipidemia

Table: 2

Fredrickson Classification of the Hyperlipidemia

Phenotype	Lipoprotein Abnormality	Result
Type I	Chylomicrons elevated	Very high TG
Type IIa	LDL elevated	High cholesterol
Type IIb	LDL and VLDL elevated	High cholesterol and TG
Type III	IDL elevated	High cholesterol and TG
Type IV	VLDL elevated	High TG, normal to slightly high cholesterol
Type V	Chylomicrons present and VLDL elevated	Very high TG and cholesterol

High Density Lipoprotein

HDLs are produced by the liver and the GI tract and by the peripheral catabolism of chylomicrons and VLDLs. HDL particles carry cholesteryl ester as their major lipid and apos A-I and A-II as their major proteins. Much of the apoprotein component of HDL is transferred in the systemic circulation to VLDLs or chylomicrons. Apo C-II an obligatory activator of LPL, is one of the apo proteins transferred by HDL. By weight, HDL particles are about 30 percent cholesterol, 45 percent protein, and 25 percent

phospholipid (predominantly phophatidyl choline). Small amounts of triglycerides are present.

HDL particles exist in several subtypes. For clinical purposes, HDL_2 and HDL_3 are the major circulating subfractions. HDL_2, which migrates with alpha mobility, is the subfraction, most closely associated with statistical protections against premature atherosclerosis. It has a density of 1.061 to 1.201 gm/ml and a diameter of 90 to 120A. HDL_3 is a smaller particle, with a density of 1.125 to 1.210 gm/ml and a diameter of 50 to 90A. Alcohol consumption increases both HDL subfractions, with a greater impact on HDL_3. Lower levels of both subfractions are associated with male gender, hypertriglyceridemia, diabetes mellitus, obesity, uremia, the use of androgens, progestins, and tobacco products; and diets rich in polyunsaturated fats but low in total fat content.

Several epidemiological studies have addressed the debate whether there is a varying clinical impact on CAD depending on the relative levels of HDL_2 and HDL_3. In males with CAD who have an associated low level of circulating HDL, both fractions of HDL are depressed with more of a decline in HDL_2.

HDL particles are thought to participate in the reverse transport of free cholesterol from peripheral tissues by way of a putative HDL receptor.

Oram and co-workers report that apos A-I and A-II interact with this receptor. This receptor mediated reverse transport could explain why patients with elevated HDL concentrations are less prone to CAD.

Another explanation for the inverse relation between HDL levels and CAD incidence may be related to the observation that most patients with low levels of HDL have elevated levels of the more cholesterol and triglyceride rich lipoproteins. In this case, Low HDL levels may serve only as a marker for other, concurrent lipid abnormalities.

Just as low levels of HDL are statistically associated with atherosclerosis, HDL is increased on a genetic basis in familial hyper apolipoproteinemia. The condition has been described as being associated with longevity.

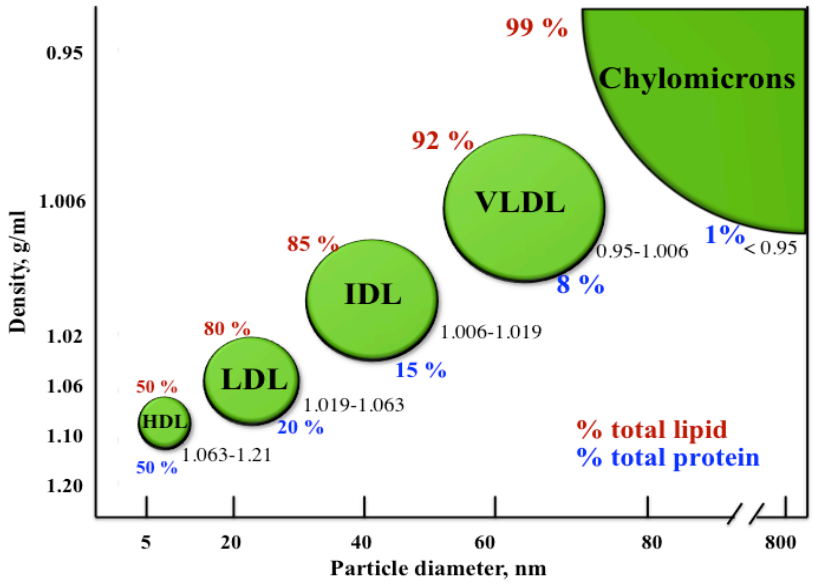


Fig.15. Comparison of density and diameter of lipoproteins

Lipoprotein (A)

Lipoprotein(a), or Lp(a), has been established as an independent CAD risk factor. The structure of Lp(a) is similar to that of an LDL molecule linked by a disulfide bridge to apoprotein(a). It has a density of l.085gm/ml and a size of 25A and it migrates in the pre beta region. Lp (a) levels range from l mg/dl to 100 mg/dl, with the largest number of values below 20mg/dl.

Although Lp(a) is structurally similar to LDL, the former appears to be regulated independently and carries an independent relation to overall coronary risk. If serum levels of both LDL and Lp(a) are elevated, the risk of CAD is markedly increased. Recent angiographic studies have documented a positive correlation between Lp(a) levels and the severity of coronary atherosclerosis.

The mechanism by which high levels of Lp(a) are related to coronary atherosclerosis is unclear. It has been suggested that because of the structural similarities of Lp(a) to plasminogen, high levels of Lp (a) may inhibit the thrombolytic activity of naturally occurring tissue plasminogen activity. Plasminogen is composed of five sequences of aminoacids rich in cysteine. Each sequence is called a kringel. Lp(a) lacks the first three kringles, but has a sequence that is highly homologous to the fourth kringle of plasminogen.

This particular sequence is repeated 37 times in the Lp(a) molecule. There is no serine protease activity in Lp (a) and no thrombolytic activity. An alternate explanation for the association between elevated Lp(a) levels and atherosclerosis is that Lp(a) may some how alter the LDL mediated delivery of cholesterol to the atherosclerotic plaque.

The control mechanisms of Lp (a) are unknown. Dietary changes that increase LDL levels do not affect Lp (a) levels. The effects of pharmacological agents are unclear, although Lp (a) has been reported to be decreased by niacin, neomycin, and stanozolol.

Lipids and Atherosclerosis

It is well established that hypercholesterolemia due to elevated blood levels of LDL is a major cause of CHD and that lowering elevated levels of cholesterol will reduce the risk of coronary disease. Inverse relationship is noted with HDL. There is also a weak correlation between plasma triglycerides and the incidence of coronary artery disease. The exact mechanism of atherogenesis is still in controversy. LDL particles which get oxidized (by natural process) may be particularly atherogenic. Receptors on the surface of macrophages within the plaque, binds and accumulates oxidized LDL. The formation of antibodies to oxidized LDL is also important in plaque formation. There is a possibility that chronic

hyperlipidemia may initiate endothelial injury which stimulates atherosclerosis. This injury favors adherence of monocytes and lymphocytes at the focus of injury, in part owing to the stimulation of endothelial cell synthesis of adhesion molecules. It induces a change in the platelet membrane composition leading to activation and increased adhesiveness of platelets. With chronic hyperlipidemia, lipoproteins accumulate within the intima at sites of endothelial injury or dysfunction.

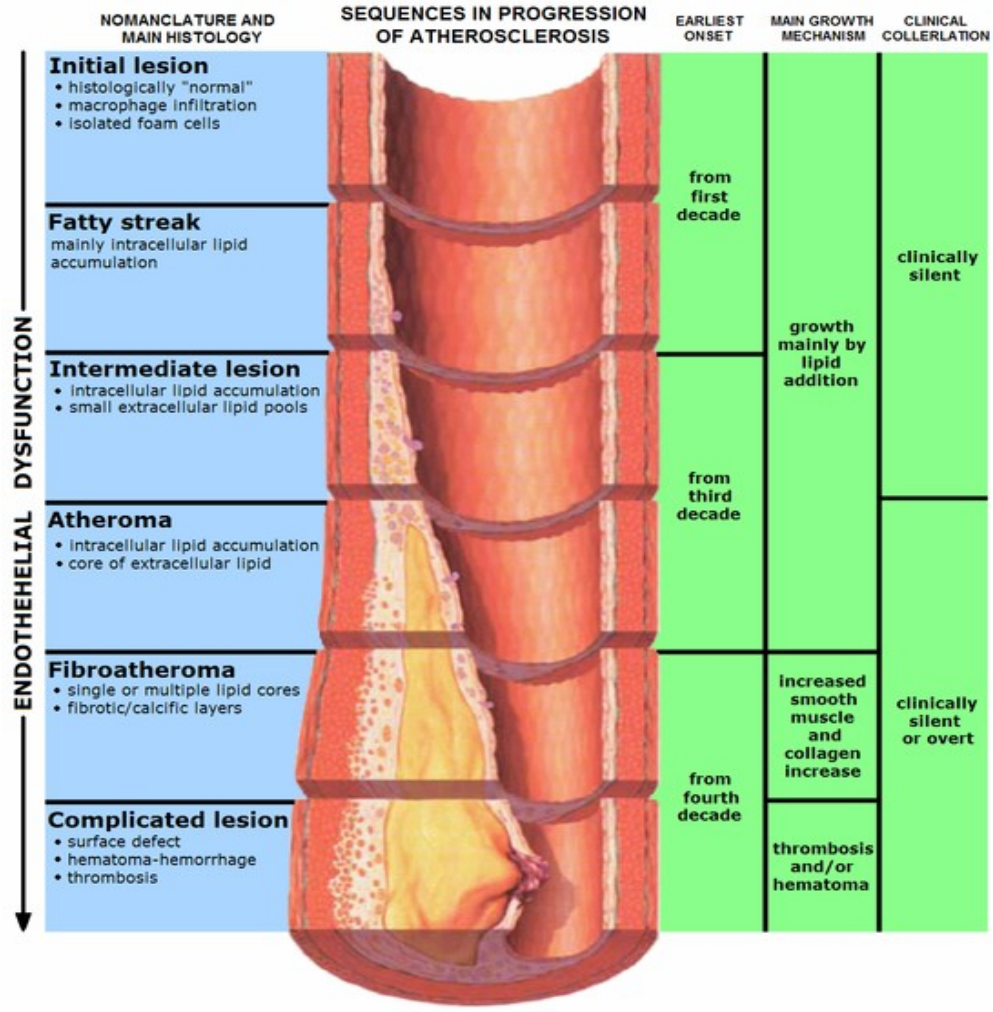


Fig. 16: Atherogenesis

A strong negative correlation has been demonstrated between plasma levels of HDL cholesterol and CHD. Most of the variability in HDL cholesterol levels reflect HDL_2 levels; plasma levels of the other major HDL subclass, HDL_3, are fairly constant both intra and inter-individually.

HDL particles secreted into the circulation by liver and cholesteryl ester enriched macrophages are complexes of apolipoproteins and phospholipids and acquire unesterified cholesterol originating in cell membranes during cell renewal or death. Their major phospholipid is phosphatidyl choline or lecithin.

Both lecithin and unesterified cholesterol serve as substrates for the cholesterol- esterifying enzyme, lecithin cholesterol acyltransferase (LCA T), which circulates with HDL in the plasma. LCAT acts on the nascent HDL particles to generate a core of cholesteryl esters and to change the structural transition to mature, spherical HDL particles. LCAT also acts on mature HDL_3 particles, i.e., after they have acquired cholesterol and lecithin not only from cell membranes but also from chylomicrons and VLDLs during lipolysis of those particles. Generation of cholesteryl esters by the LCAT reaction leads to enlargement of the small HDL_3 particles such that they are converted into the larger HDL_2 particles. Formation of HDL_2 increases the cholesterol-carrying capacity of HDL, and HDL cholesterol

levels raise, this drives the process termed “reverse cholesterol transport”, in which HDL returns cholesterol from peripheral tissues to the liver for excretion into the bile.

The cholesterol esters of HDL, particularly of HDL_2 formed de novo by the LCAT action, need not remain within the HDL core. They can be transferred from HDL to the triglyceride rich lipoproteins, i.e., chylomicrons and VLDL in exchange for triglyceride molecules. This hetero exchange of insoluble cholestery lesters and triglycerides between HDL and triglyceride rich lipoproteins is catalyzed by the action of cholesterol ester transfer protein.

The transferred triglycerides are hydrolyzed from the HDL core by hepatic lipase, located in the endothelial cells of the liver. Only cholesteryl esters those that were not exchanged for triglycerides remain in the core. Hence HDL_2 particles are converted back into the smaller HDL_3 particles. This mechanism is the basis for the well-established clinical observation that, individuals with permanent or temporary hypertriglyceridemia (due to increased levels of VLDL or due to accumulation of chylomicrons in the course of postprandial lipemia) have low $HDL_2$and low HDL cholesterol levels.

The cholesteryl esters transferred from HDL and the triglyceride rich

lipoproteins remain with the latter particles along their lipolytic cascades and the endocytotic pathways of their remnants (i.e., the LDL receptor and the scavenger pathways). Thus, transfer of cholesteryl esters from HDL to triglyceride rich lipoproteins may contribute to the atherogenic potential of chylomicrons and VLDL, in that "good" cholesterol is turned into "bad" cholesterol.

Although there is no clinical trial directly demonstrating the benefit of increasing HDL cholesterol alone, Helsinki Heart Study results suggests that increasing HDL add benefit to lowering LDL in CHD risk reduction. Similarly, data from the Prospective Cardiovascular Muenster (PROCAM) Study have shown that the combination of LDL elevation, hypertriglyceridemia, and low HDL confers greater risk of CHD than elevated LDL alone.

There are several causes for low serum HDL cholesterol levels. Heavy cigarette smoking is a documented cause. Obesity is an apparent association, as is a sedentary lifestyle. Hypertriglyceridemia is frequently associated with very low HDL levels. Certain drugs can also have an appreciable depressing effect on HDL. Finally, there is evidence that isolated low HDL cholesterol, termed hypoalphalipoproteinemia, may be genetically transmitted in an autosomal dominant fashion. Patients with this disorder have a normal lipid

profile other than the low HDL but an apparently increased risk for atherosclerosis.

There commended methods for improving HDL cholesterol values are non- pharmacologic; smoking cessation, weight reduction, regular and vigorous exercise and alteration of offending drugs, if possible. As yet, there is no direct evidence that drug- induced increases of low HDL cholesterol in the setting of normal LDL cholesterol and triglyceride levels are beneficial in CHD risk reduction. HDL cholesterol levels can be increased with gemfibrozil (the agent used in the Helsinki study) or nicotinic acid.

The possibility that chronic hyperlipidemia, and particularly hypercholesterolemia, may itself initiate endothelial injury has enticed many investigators. Much evidence suggests that hypercholesterolemia has a variety of adverse effects.

It increases the cholesterol- phospholipid ratio of endothelial cell membranes, rendering them more rigid and less able to maintain their normal intercellular associations, potentially increasing permeability and in effect causing a subtle form of endothelial injury.

It favours the adherence of monocytes and lymphocytes to the focus of injury, in part owing to the stimulation of endothelial cell synthesis of adhesion molecules.

It induces changes in platelet membrane composition, leading to activation and increased adhesiveness of platelets.

With chronic hyperlipidemia, lipoproteins accumulate within the intima at the site of endothelial injury or dysfunction. Although significant amounts may enter the arterial wall, there is some concurrent efflux, perhaps mediated by HDL.

Most important, it provides the opportunity for oxidation of lipoproteins, yielding modified LDL.

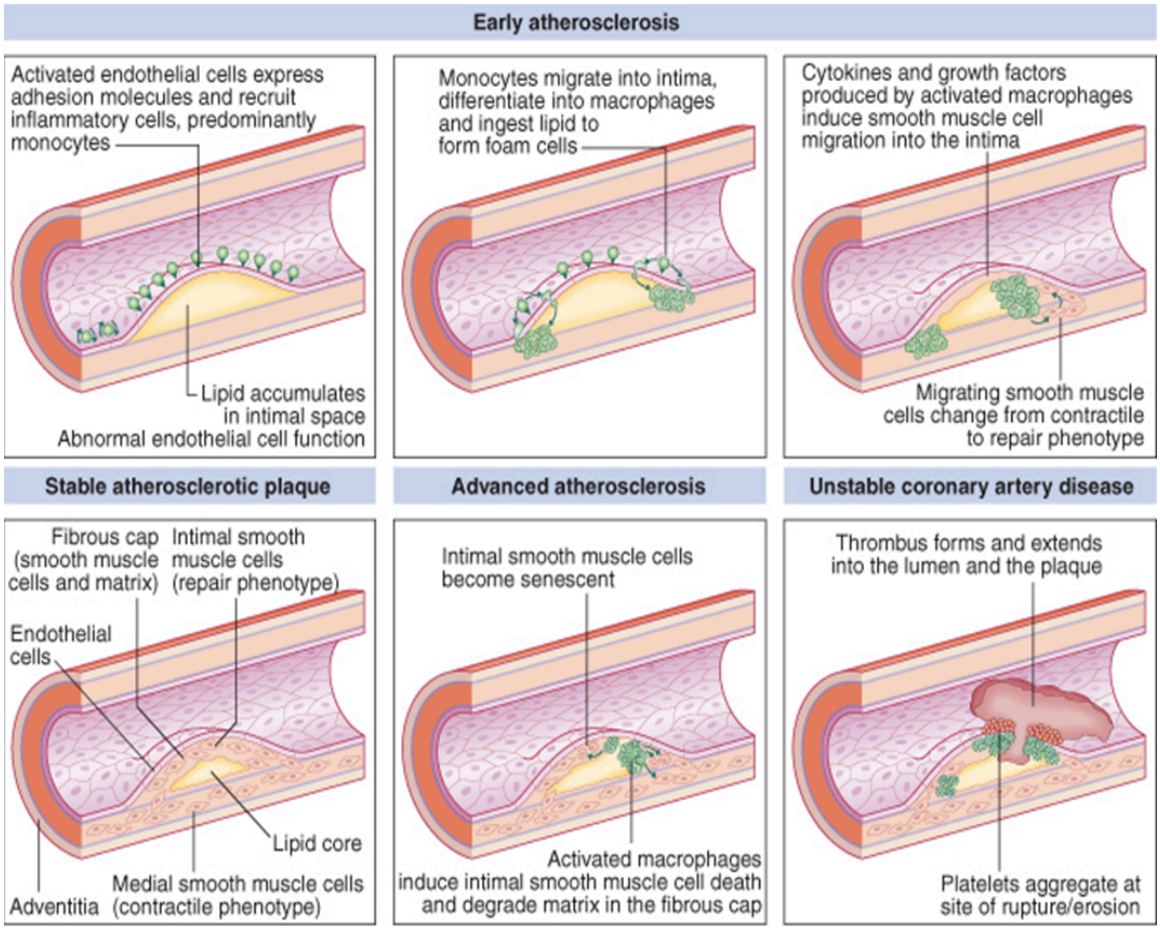


Fig. 17: Stages of atherosclerosis

Apoproteins

Apoproteins are key lipoprotein components that serve both as enzymatic co- factors and as recognition elements that bind to specific receptors on peripheral tissues, including the vascular endothelial cells. It is the apo E component of the chylomicron remnant, for example, that is recognized by receptors on the hepatocyte. The apoproteins are distinguished alphabetically and numerically as apo A-I through apo E.

A great deal of research has been conducted in the use of apoproteins as CAD markers. Some investigators have found that the concentration of apo A-I and apo B-100 are better predictors of CAD than are measurements of total plasma lipids or lipoproteins. In one study, apo A-I was the best predictor of atherosclerotic risk in patients undergoing coronary arteriography. In this study, higher levels of apo A-I were associated with a deceased prevalence of obstructive coronary lesions. Indeed, apo A-I was found to be a better CAD predictor than either total cholesterol or HDL.

Apoprotein A

Apo A-I, the prototype of apo A, is a major protein in HDL and also is seen in chylomicrons. It has a molecular weight of 28,000 and is synthesized in the GI tract and liver. Its specific regions called amphipathic helices are enriched with charged amino acids that form areas of polar and non-polar

residues. Apo A-I functions as the activator of lecithin cholesterol acyl transferase (LCAT), and also has been found as a degradation product in amyloid fibrils.

Apo A-II is a minor constituent of HDL and does not appear to be present in all species. Human apo A-II is of hepatic origin and consists of two identical chains attached by a single disulfide linkage. Apo A-II may be an activator triglyceride and phospholipid while utilizing HDL_2 as its preferred substrate. Apo A-IV is synthesized in the gut and is present in HDL, chylomicrons and as a free protein. Its molecular weight is 46,000, and its structure is helical. Although its function is not known, it also may be an activator of LCAT.

The genetic codes for apos A-I, A-IV and C-III are close together on the long arm chromosome 11.Combined A-I/C-III deficiency are associated with severe premature atherosclerosis.

Apoprotein B

Apo B occurs in two forms. Apo B-48 is synthesized by the small intestine, and apo B-100 is secreted by the liver. Apo B-48 is present on the surface of chylomicrons and chylomicron remnants. Apo B-100 is found in VLDL, IDL and LDL. Apo B-I00 is the primary apoprotein of LDL and accounts for 25 percent of its weight. It also is the recognition site

for the LDL or apo B/E receptor on cell surfaces. It has recently been determined that a single gene regulates the synthesis of both apo B-48 and apo B-100. The gene for apo B-I00 has been localized to chromosome 2 and exists as a 40 kilobase structure. The structure in the amino acid sequence of human apo B-100 and the corresponding cDNA messenger have recently been determined. A unique editing mechanism introduces a stop codon into the mRNA for apo B by means of a single base change. This allows the biosynthesis of two proteins from a single gene and mRNA, with either apo B-1 00 or apo B-48 being synthesized.

Apoprotein E

Most apo E is synthesized in the liver. However, other tissues, including the small bowel, kidney, adrenals, and the cells of the reticuloendothelial system, have the ability to synthesize this apoprotein. Apo E accounts for about 15 percent of the protein content of VLDL, 7 percent of the protein content of chylomicron remnants and 2 percent of the protein content of HDL. It can be recognized by the LDL or apo B/E receptor and by specific apo E receptors in the liver whose function appears to be the removal of chylomicron remnants. Apo E is polymorphic and contains three major alleles; apos E2, E3, and E4. These respective alleles are present in about 10 percent, 76 percent, and 13 percent of whites. Their

various combinations result in homozygtes for apos E2/2, E4/4.

Also, apos E2/3, E2/4 and E3/4 exist in the heterozygous state. The polymorphism of apo E has been determined on a molecular basis and results from the substitution of an amino acid at residues 112 and 158 in the protein. About 90 percent of the patients with type III hyperlipoproteinema (HLP) are homozygous for the E2/2 phenotype. This disorder is characterized by hypercholesterolemia, hypertriglyceridemia and IDL or VLDL particles abnormally enriched in cholesterol. These particles have beta electrophoretic mobility and are termed beta-VLDLs. Premature coronary and peripheral vascular disease is characteristically associated with type III HLP. Type III HLP is also by the delayed clearance of chylomicron remnants in the serum due to impaired binding of these remnants to the lipoprotein receptors in isolated cells. Because the E2/2 genotype occurs in 1 percent of the population, and type III HLP is rare, a second abnormality must be present.

Apo E isoforms may account for as much as 15 percent of the variability of cholesterol and LDL levels in the population. Also, recent Finnish studies suggest that E4 may be associated with increased cholesterol absorption in the GI tract. In the Prospective Cardiovascular Muenster (PROCAM) study, E2 was associated with lower cholesterol levels and E3

or E4 with higher levels of total cholesterol and LDL, in populations with and without CAD.

Table: 2

SUMMARY OF APOPROTEINS

NAME	LIPOPROTEIN	FUNCTION
Apo A-I	HDL, Chylomicrons	Structural; activator of LCAT enzyme
Apo A-II	HDL, Chylomicrons	Structural
Apo A-IV	HDL, Chylomicrons, VLDL	Unknown
Apo B-IOO	LDL, VLDL	Structural; Synthesis and secretions of VLDL; bind to LDL receptors (B/E)
Apo B-48	Chylomicrons	Structural; Synthesis and secretion from intestine
Apo C-I	HDL, Chylomicrons, VLDL	Activator of LCAT
Apo C-II	HDL, Chylomicrons, VLDL	Activator of Lipoprotein lipase
Apo C-III	HDL, Chylomicrons, VLDL	Stabilizes surface; provides negative charges
Apo D	HDL, Chylomicrons	Cholesterol ester exchange
Apo E	HDL, VLDL, Chylomicrons	Binds to receptor on cell membrane of liver (E and B/E) and macrophage.

SMOKING AND CORONARY HEART DISEASE

Coronary heart disease is common in both the developed countries and developing countries. It is estimated that approximately 1.5 million infarcts occur in USA alone. Hyperlipidemia a major risk factor for atherosclerosis is characterized by raised levels of lipids (triglyceride cholesterol) and lipoproteins (low density lipoproteins) and very low-density lipoproteins. These factors vary among various socioeconomic states.

Cholesterol was postulated to be related to atherosclerosis when it was found to be a major component of advanced atherosclerotic plaques. The association between elevated serum cholesterol and atherosclerotic disease was first reported in 1930. Subsequent large epidemiologic studies such as the seven countries study and the Framingham heart study confirmed the relationship between serum cholesterol and CAD. In the multiple risk factor intervention trial (MRFIT), the relationship between serum cholesterol level and subsequent CAD was found to be continuous, graded and strong.

The lipid research clinics prevalence study demonstrated, in a 10-year follow up, that low-density lipoprotein cholesterol was strongly associated with CAD death in men with or without CAD.

In John Hopkins precursors study, cholesterol levels determined in young men in their early twenties were predictive of risk of developing

CAD 3-4 decades later. The concept of plaque stabilization in which cholesterol reduction alters vulnerable plaque in a way that may not substantially change its angiographic stenosis is not well understood. It is thought to be due to lipid depletion of the lesion core, stabilization of endothelium, inhibition of platelets or effects on vascular smooth muscle cells. Helsinki heart study has shown reduced cardiovascular mortality with cholesterol reduction.

Cigarette smoking acts both independently and synergistically with other risk factors like hypertension and hypercholesterolemia. Mortality from coronary heart disease is substantially higher in cigarette smokers than non-smokers. It is generally believed that harmful cardiovascular effects of smoking are partly caused by changes in lipid metabolism.

Gofman JW et al, pioneers in the study of the relationship of smoking, serum lipoproteins and heart diseases found a strong relationship between themselves as well as with coronary artery disease.

According to the studies of Albany and Framingham, the incidence of myocardial infarction in heavy smokers is three times more than in non-smokers. In Framingham offspring study, Wilson PW, Anderion KM et al studied patients from 1972-1974. The patients being the off springs of the original participants in the Framingham Heart Study, they underwent a

baseline examination for standard cardiovascular risk factor. The off springs were 30-59 years old and free of coronary heart disease. They were followed for 12 years during which time 156 out of 1663 men and 55 out of 1714 women developed CHD. The result of that multivariate proportional hazard model was that, coronary heart disease is significantly related to age, reduced HDL and the amount and duration of tobacco smoking. Blood sugar levels and LDL values were strongly related with coronary heart disease in males, but less significantly in females, while triglycerides and VLDL were not associated with coronary heart disease after adjustment for high density lipoprotein cholesterol and glucose.

Castelli WP et al, who reviewed with the Framingham study on coronary heart disease have shown that life style, particularly diet, smoking and alcohol consumption have a great impact on the incidence of coronary heart disease. Blood lipoproteins, rather than total cholesterol have been found to be more accurate predictors of coronary heart disease. Blood triglycerides previously considered to have little bearing on a coronary heart disease risk was found to have a negative impact in many cases.

Knutsen SF et al in Tromso Heart Study on the risk for coronary heart disease in wives of coronary high risk men found a significantly higher total cholesterol ratio, higher body mass index and higher coronary risk

scores compared to the age matched married women in general population. Furthermore, higher proportions of these individuals were daily cigarette smokers and had received fewer years of education. The members of the same household of a person with an increased risk for CHD also had increased risk, probably due to shared life style.

In a study of all patients admitted to the coronary care unit of St. Vincent's Hospital in Dublin, Ireland, which numbered 978 with a first documented myocardial infarction, a study was made to detect smoking related differences in clinical profile and in hospital outcome. The distribution of infarct size differed significantly between smokers and non-smokers. Smokers had a higher cardiac enzyme concentration. The study was conducted between 1980-85. Tomono, Sand Ohshima S reviewed the histories and laboratory data of 67 young patients with ischemic heart disease under the age of 40 years. The risk factor in 13 of them was heavy smoking. But it came second to hypercholesterolemia patients, who number 23. The results indicated that hypercholesterolemia is the most important risk factor for ischemic heart disease in young patients, although other accepted risk factors like hypertension, diabetes and cigarette smoking were associated with the development of the disease.

Fujiwara et al, conducted a study, to assess the metabolic risk factor in

normolipidemic patient with angiographically defined CHD. In the CAD group cumulative lifetime tobacco consumption was higher and HDL cholesterol concentration was lower than that in those without coronary artery disease group.

Smoking accelerates myocardial infarction not only by accelerated development of atherosclerosis but also by augmenting the existing thrombotic and vasospastic effect. The arrythmogenic effects of smoking increases the risk of threatening arrhythmias and sudden cardiac death and myocardial infarction occurred in young smokers with nearly normal or normal coronary arteries. Smokers who undergo coronary artery bypass surgery have increased perioperative mortality compared to non-smokers. Cigarette smoking may produce an imbalance between myocardial O_2 demand and supply.

SMOKING AND PERIPHERAL ARTERIAL DISEASE:

Peripheral arterial disease (PAD) in most of the developed countries is due to atherosclerosis and so tobacco smoking is strongly associated with PAD. Most of the serious manifestations of PAD are due to rupture of the plaques. In UK about 25% of the middle aged persons are having PAD. But only one fourth of them are having the symptoms. The manifestations vary

according to the site of involvement, presence of collateral supply, speed of onset and mechanism of injury.

CHRONIC LOWER LIMB ARTERIAL DISEASE:

PAD affects the lower limbs more often than the upper limbs(around 8 times more).

The presentation of leg ischemia may be as:

1. Intermittent claudication (IC)
2. Critical limb ischemia (CLI)

The severity of limb ischemia is assessed by

1. Clinical examination
2. ABPI (ankle brachial pressure index) – is the ratio between highest systolic ankle and brachial blood pressure.
    a. Normal>1.0
    b. I.C- 0.5-0.9
    c. C.L.I. <0.5

INTERMITTENT CLAUDICATION:

It is the pain in the muscles of leg which occurs while walking, and is due to ischemia. As the superficial femoral artery is the one which is commonly involved, the pain is commonly present in the calf muscles. If the disease involves iliac arteries the pain occurs in the buttock or thigh muscles.

Normally the pain occurs after a particular claudication distance. When walking is stopped the pain gets subsided immediately. Then the pain recurs when walking is resumed. Due to progression of the disease and development of collateral blood supply, mostly the patients will be experiencing a cyclical pattern of exacerbation and then resolution of symptoms.

Around 4.5% of men in middle age will experience intermittent claudication. If the patients comply well with medical management, only 0.5-2% will deteriorate and go for amputation or revascularization. But the annual mortality in these patients is more than 6%, which is 3 times more than the non-claudication population. Because this claudication pain is almost always associated with diffuse atherosclerosis, these patients face death due stroke or myocardial infarction.

The best medical therapy includes mainly supervised exercise therapy. The use of peripheral vasodilators improves the distance of walking. Other interventional procedures are:

1. Stenting
2. Endarterectomy
3. Angioplasty
4. Bypass surgeries

These procedures are considered in patients who are disabled severely and not improved symptomatically even after 6 months of best medical therapy.

CRITICAL LIMB ISCHEMIA:

- Rest or night pain will be present.
- Associated with loss of tissue (gangrene or ulceration).
- Persisting > 2 weeks.
- With ankle B.P. of less than 50mmhg

SUBCRITICAL LIMB ISCHEMIA:

- Only rest pain is present
- Ankle B.P. more than 50mmhg

Severe limb ischemia includes both critical and subclinical limb ischemia. In intermittent claudication plaque will be present in a single segment only. In severe limb ischemia plaque will be present in multiple levels. These patients with severe limb ischemia are at higher risk of losing limb when compared to those with intermittent claudication. But treatment in these patients is difficult as they almost have extensive end stage disease and they also have associated multisystem involvement by atherosclerosis.

BUERGERS DISEASE:

This is an inflammatory obliterative arterial disease. It usually affects male smokers of young age. It typically involves distal arteries and present with claudication of feet and rest pain of fingers. Pulses are absent in wrist and ankle but present in brachial and popliteal vessels. Superficial thrombophlebitis also occurs as the disease involves veins. The disease mostly remits on cessation of tobacco smoking. If these patients don't stop smoking, the disease progresses to the level of amputation. Prostaglandin infusions and sympathectomy might be useful.

SMOKING AND RESPIRATORY DISEASES:

89% of chronic obstructive pulmonary disease is due to tobacco smoking. Within first few years of starting smoking, the smokers manifest inflammatory changes in their smaller airways. But lung function tests during this period wont predict the evolution to chronic airflow limitation. In chronic smokers pathophysiologic changes start developing after 2 decades and these changes in lung progress depending on the time duration and severity of smoking. More than 70% of the smokers will be having a productive cough due to the presence of chronic mucous hyperplasia in large airways. Narrowing of the smaller airways due to chronic inflammation and enzymatic degradation of walls of alveoli leads to emphysema. This

produces expiratory airflow limitation, which leads to clinical symptoms in around 22% of smokers. In smokers of young age group small airway related changes gets reversed within one to two years of smoking cessation. A small improvement in expiratory airflow changes is observed in smokers who have developed chronic airflow limitation after smoking cessation. But the important change after cessation is deceleration of declining lung function rather than returning to normal.

SMOKING AND CANCERS:

Smoking is associated with cancers of oropharynx, nasopharynx, hypopharynx, oral cavity, larynx and lungs. It is also associated with cancers of oesophagus, stomach, pancreas, body and pelvis of kidney, urinary bladder, ureters, and cervix. It also increases the risk of leukemias of myeloid series. There is also increased risk of developing hepatoma, breast cancer (premenopausal), and colorectal cancers. It is not found to be related with postmenopausal breast cancers. In postmenopausal smokers there is a reduced risk of developing cancers of endometrium. There is a synergesic effect of alcohol with smoking in the development of malignancies of oesophagus, oral cavity and lung. Occupational exposures like radon and asbestos act synergistically with tobacco to increase the risk of developing cancers of lung. Smoking cessation produces a reduced risk of smoking

related malignancies. But even after 2 decades of stopping smoking there is modest increased risk of lung malignancies.

SMOKING AND PREGNANCY:

The maternal problems related to smoking are increased spontaneous abortions, placenta previa, placental abruption and increased risk of premature rupture of membranes. Infant related problems are preterm delivery, small for gestational age, increased perinatal mortality and increased risk of respiratory distress syndrome. These children will experience a developmental delay in the first few years of their life.

OTHER CONDITIONS ASSOCIATED WITH SMOKING:

- Osteoporosis
- Senile cataracts
- Peptic ulcers
- Premature menopause
- Macular degeneration
- Skin wrinkling
- Male impotence
- Cholecystitis in women

- Gall stones

ENVIRONMENTAL TOBACCO SMOKE:

Non-smokers who have prolonged exposure to environmental tobacco smoke are at increased risk of developing coronary artery disease and lung cancers. Among children there is increased risk of bronchial asthma, chronic otitis media, and respiratory infections. There is also increased risk of premenopausal breast cancer.

CESSATION OF SMOKING:

- More than 75% of smokers like to stop smoking.
- Every year 1/3rd of smokers try to quit the habit.
- About 85% of the unassisted attempts to quit fail.
- The process of cessation is a cyclical one that is, the person tries to quit smoking several times and fails repeatedly before succeeding finally.
- Interventions for quitting smoking should be clinician based & the smokers should be encouraged to try quitting.
- Rather than advising abrupt cessation in the first visit, various forms of cessation assistance should be advised with each new attempt to quit smoking.

- If the advice to quit smoking is given by the physician at the time of any acute illness it will be a strong trigger, and thereby most of the persons will make an effort to stop smoking.
- Various nicotine replacement products are available. They are:
    - Nicotine gums
    - Nicotine lozenges
    - Nicotine patches
    - Nicotine oral inhalers
    - Nicotine nasal inhalers
- The products can be used up to 6 months.
- Antidepressant- Bupropion is found to be effective.
- A partial agonist of nicotinic acetylcholine receptor, varenicline is also found to be effective. Psychiatric symptoms like suicidal ideations are found to be associated with the use of this drug.
- Two weeks before the planned quitting date pretreatment with varenicline or antidepressants is adviced.
- Nicotine replacement products can also be used in pretreatment. For more intense smokers higher doses are advised.
- In patients who failed first line treatment, nortriptyline or clonidine may be useful.

- A combined approach with advice, counseling and pharmacological assistance mostly makes the cessation successful.

PREVENTION OF SMOKING:

- Most of the smokers start this habit in their teen age. The reasons behind this are:
    - Parental cigarette smoking.
    - Advertising and promotional activities for smoking tobacco.
    - Easy availability of cigarettes.
    - Acceptability of smoking by the society.
    - Adult imitating behavior and need for enhanced self image.
- So prevention:
    - Must be started earlier even in primary school years.
    - Physicians treating teen agers should enquire about this habit and explain them the fact that most adults and adolescents don’t smoke.
    - Physicians should explain them the harmful effects and additive nature of tobacco in all forms.

4. METHODOLOGY

This study was conducted among 200 patients who attended Government Rajaji hospital, Madurai., out patient department for various ailments between July 2014 to September 2014.

INCLUSION CRITERIA:

1. Age: 20-50 years
2. 100 males who never smoked.
3. 100 males smoking for 5 or more years, divided into three groups depending on intensity of smoking as follows
    a. Mild Smokers (1 – 10 Cigarettes / Bidis per day)
    b. Moderate Smokers (11 – 20 Cigarettes / Bidis per day)
    c. Severe Smokers (> 20 Cigarettes / Bidis per day)

EXCLUSION CRITERIA:

1. Obese
2. Those on diet restriction.
3. Those on drugs known to alter lipid profile.
4. Those with history of alcohol intake.

5. Those with diseases known to alter lipid profile like diabetes mellitus,
6. Hypothyroidism, renal failure.
7. Those with hypertension and coronary artery disease.
8. Those with family history of dyslipidemia.

Since smoking is less prevalent among women in our country this study was conducted only in men.

Blood Sampling and Laboratory Evaluation

The blood samples for analysis were taken at least after minimum of 12 hours of complete fasting. The subject were asked to have a light, fat free diet on the day prior to sampling.

The venepuncture was done in the cubital fossa, Tourniquet was used but was released just before sampling to avoid artifactual increase in the concentration of serum lipids.

About 10 ml blood was drawn using perfectly dry and sterile disposable syringes. The serum was separated within 2 hours of collection to prevent artifactual changes in concentration of HDL. The sample were analyzed the same day or within 48 hours.

The lipid and lipoprotein assay was done using the Dr. Lange LP 700 equipment.

Description of the Laboratory System LP 700: (Dr Lange)

The laboratory system LT 700 consist of the following single instruments:

- LP 700 Photometer (1)
- LQV 016 Universal Thermostat (2)
- LTV 015 Cuvette changer (Rack) Thermostat (3)
- LQV 018 Suction Device (4)

The filters of the wavelengths 340 nm, 405 nm, 492 mm, 546 nm, 578 nm belong to the LP 700 and are automatically recognized by it. The following standard wavelengths 520 nm, 623 nm and 800 nm are available additionally.

The cuvette compartment accepts any 10 mm square cuvette as well as the Dr. Lange Cuvette Test.

The minimum volumes are:

- 500 micro liter for SM square cuvette and
- 450 micro liter for SM Suction cuvette
- The filling volume of the Dr. Lange Cuvette Test is prefixed.

The methods adopted for estimating the various lipid fraction is as follows:

1) Serum cholesterol Estimation:

This was done on a method based on CHOD-PAP method (Boehringer Mannheimn GmbH diagnostic catalogue No. 237574).

Method: Enzymatic colorimetric method.

Test Principle:

Cholesterol esterase (CHE) hydrolyses cholesterol ester. Free cholesterol is oxidized by the cholesterol oxidase (CHO) to Cholest-4-En-3-one and hydrogen peroxide. Hydrogen peroxide formed reacts with 4-amino antipytine and phenol in the presence of peroxidase (POD) to produce pin colored dye.

$$\text{Cholesterol esters} + H_2O \xrightarrow{\text{Cholesterol esterase}} \text{cholesterol} + \text{RCOOH}$$

$$\text{Cholesterol} + 02 \xrightarrow{\text{Cholesterol oxidase}} \text{cholest} - 4 - \text{en} - 3 - \text{one} + H_2O_2$$

$$2H_2O_2 + 4 - \text{a min ophenazone} + \text{phenol} \xrightarrow{\text{peroxide}} 4 - (\text{P} - \text{benzoqui none} - \text{monoi min o}) - \text{Phenazone} + 4H_2O_2$$

Normal cholesterol value in serum: 150-240 mg/dI

2) Serum Triglyceride Estimation:

Triglyceride concentrations were determined by enzymatic colorimetric test (Boehringer Mannheim GmbH Cat. No. 701882) as per the details shown in the system specific working instructions supplied with the kits.

Method:

Enzymatic hydrolysis of triglycerides with subsequent determination of the liberated glycerol by colorimetry

Test Principle:

Triglycerides are hydrolyzed by lipase to glycerol and free fatty acids. Glycerol is phosphorylated by ATP in the presence of glycerol kinase to glycerol phosphate (G-3-P) which is oxidized by the enzyme glycerol 3-phosphate oxidase (G-P-O) producing hydrogen peroxide. Hydrogen peroxide so formed reacts with 4-aminoantipytine/3, 5 dichloro-2-hydroxy benzene sulfuric acid to give a red colored complex which is read at 510nm (500- 50 nm)

$$\text{Triglycerides} + 2\,H_2O_2 \xrightarrow{\text{lipase}} \text{glycerol} + 3\,\text{RCOOH}$$

$$\text{Glycerol} + \text{ATP} \xrightarrow[Mg2+]{\text{GK}} \text{glycerol} - 3\,\text{phosphate} + \text{ADP}$$

$$\text{Glycerol} - 3 - \text{phosphate} + O_2 \xrightarrow{\text{GPO}} \text{dihydroxyacetone phosphate} + H_2O_2$$

$$H_2O_2 + 4 - \text{a min ophenazone} + 4 - \text{cholorophenol} \xrightarrow{\text{peroidase}}$$
$$4 - (\text{p} - \text{benzoquinone} - \text{monoi min o}) - \text{phenazone} + 2H_2O + \text{Hcl}$$

Clinical Interpretation:

Normal range: 74-172 mg%

3) HDL Cholesterol Estimation:

This involves two steps-precipitation and cholesterol estimation of the HDL – fraction by a modification of the method described by Burstein et al.

Test Principle:

Chylomincrons, VLDL (very low-density lipoprotein), and LDL (low-density lipoprotein) are precipitated by adding phosphotungstic acid and magnesium ions to the sample. Centrifugation leaves only the HDL (high – density lipoproteins) in the supernatant; their cholesterol content is determined enzymatically. (Boehringer Mannheim GmbH Cat. 543004).

In patients with high triglycerides values: the HDL cholesterol estimation was done after dilution of serum (1:1) with isotonic saline and the resultant cholesterol value of HDL-cholesterol was multiplied by 2. This was done to prevent the erroneous values of HDL-cholesterol due to impaired sedimentation of the precipitate in a serum with high triglycerides concentration.

Clinical interpretation

Normal range – 35 to 55 mg %

It is considered as a risk factor if HDL-cholesterol level was below 35 mg %

4) LDL Cholesterol Estimation:

LDL – cholesterol was calculated by using a standard WHO approved formula based on the total cholesterol, triglyceride and the HDL – cholesterol values.

LDL cholesterol = Total cholesterol – (HDL + triglyceride/5)

Triglyceride/5 indicates the cholesterol in VLDL fraction and was used when the TG levels were below 400 mg/dI

5. STATISTICAL ANALYSIS

The mean levels of various variables were correlated with basal reference for normal individuals. Relevant statistical methods like student ‘t’ test and whenever required Mann-Whitney test was used to see the significance of difference in mean values between groups and to know there correlation between inter and intra group variation.

Ethical clearance was obtained from the ethical committee.

Table: 4

Comparison of Lipid Profile among Smokers and Non Smokers

Lipid Profile	Non-Smokers (Mean ± Std)	Smokers (Mean ± Std)	P-Value
TC	161.18±26.77	191.96±31.37	< 0.001
TG	103.58±26.26	164.29±28.95	< 0.001
HDL	49.58±8.57	44.72±9.96	= 0.002
LDL	82.34±16.57	103.08±18.66	< 0.001
VLDL	21.69±6.42	29.02±8.98	< 0.001

Figure: 18

Comparison of Lipid Profile among Smokers and Non Smokers

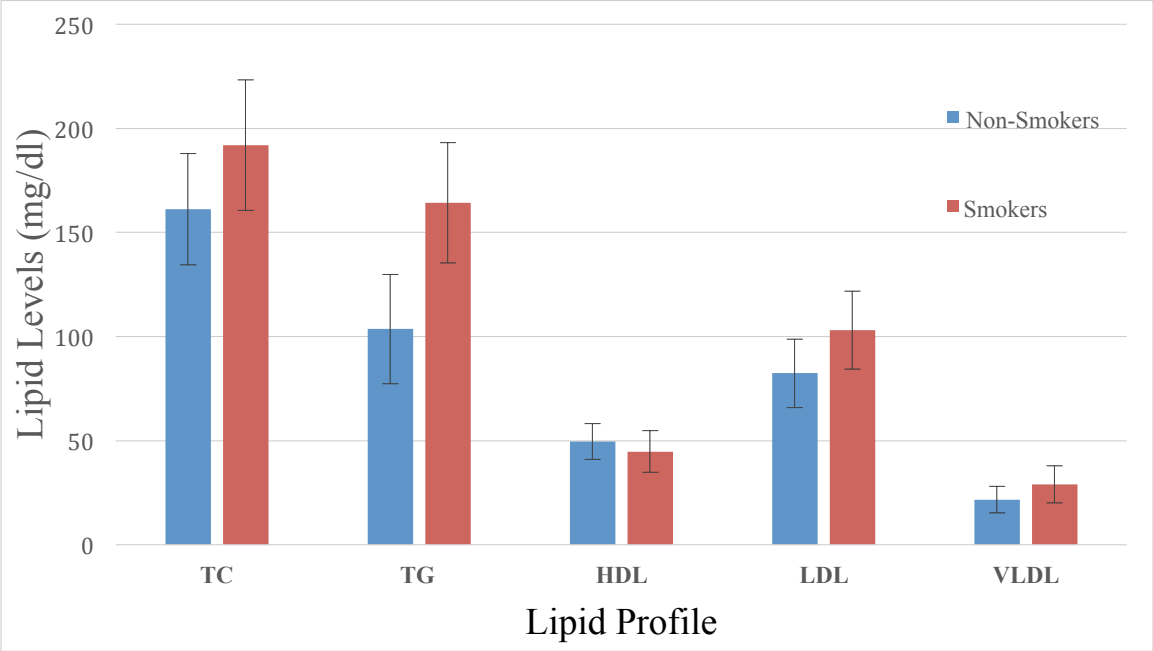


Table: 5

Comparison of Lipid Profile depending on Intensity of Smoking

Lipid Profile	Smokers		
	Mild	Moderate	Severe
TG	183.06±33.38	194.09±29.39	209±29.15
TC	155.06±31.8	166±26.63	176.5±25.62
HDL	48.03±8.65	45.06±10.26	42±9.67
LDL	97.12±13.05	105.58±20.07	114±20.41
VLDL	23.15±7.67	32.73±6.67	34±9.27

Figure: 19

Comparison of Lipid Profile depending on Intensity of Smoking

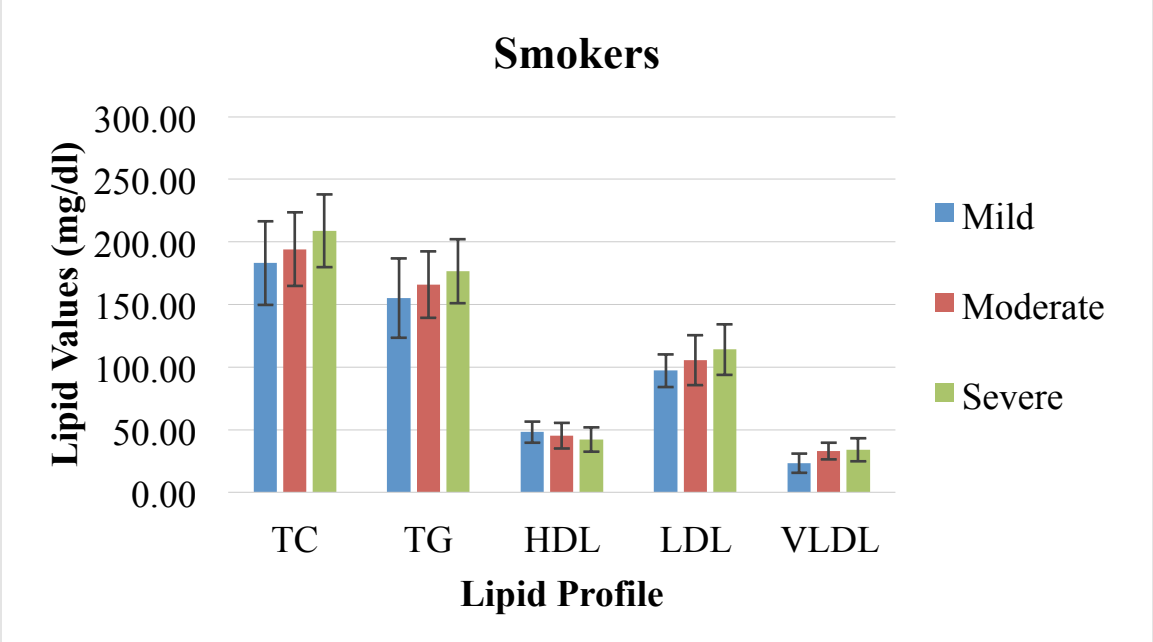


Table: 6

Comparison of Lipid Profile among Non Smokers and Smokers

(with respect to intensity of smoking)

		Non-Smokers Mean±Std	**Smokers Mean±Std**	**p value**
TC	Mild	161.18±26.77	183.06±67	< 0.05
	Moderate	161.18±26.77	194.09±29.39	< 0.001
	Severe	161.18±26.77	209±29.15	< 0.001
TG	Mild	103.58±26.26	155.06±68	< 0.001
	Moderate	103.58±26.26	166±26.63	< 0.001
	Severe	103.58±26.26	176.5±25.62	< 0.001
HDL	Mild	49.58±8.57	48.03±8.79	< 0.5
	Moderate	49.58±8.57	45.06±10.26	<0.07
	Severe	49.58±8.57	42±9.67	< 0.05
LDL	Mild	82.34±16.57	97.12±13.26	< 0.001
	Moderate	82.34±16.57	105.58±20.07	< 0.001
	Severe	82.34±16.57	114±20.41	< 0.001
VLDL	Mild	21.69±6.42	23.15±7.65	< 0.001
	Moderate	21.69±6.42	32.73±6.67	< 0.001
	Severe	21.69±6.42	34±9.27	< 0.001

Figure: 20

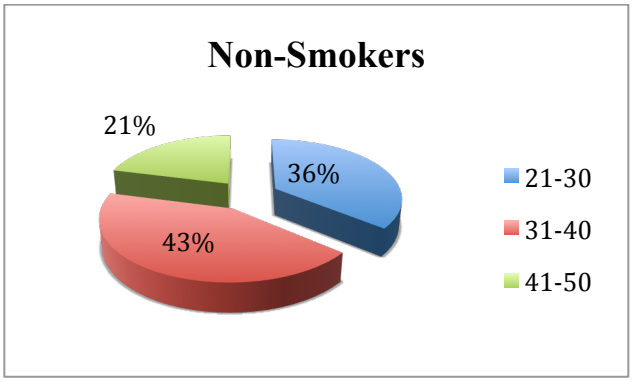
Comparison of Lipid Profile among Non Smokers and Smokers

(With respect to intensity of smoking)

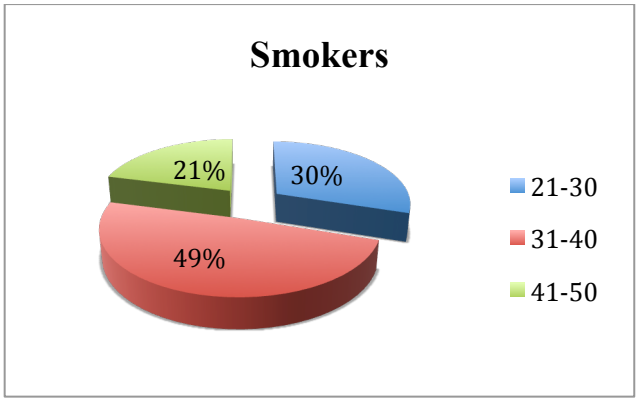
Table: 7

Age Distribution of the Study Group

Age in Years	No. of Non-Smokers	No. of Smokers
21-30	36	30
31-40	43	49
41-50	21	21



(a)



(b)

Figure: 21.

Table: 8

Comparison of Total Cholesterol among Smokers and Non-Smokers

(Age wise)

	Non-Smokers		Smokers	
Age in Years	No. of Persons	TC	No. of Persons	TC
21-30	36	156.64±22.58	30	197.17±32.59
31-40	43	163.84±29.21	49	191.45±29.72
41-50	21	163.52±27.11	21	185.71±32.08

Figure: 22

Comparison of Total Cholesterol among Smokers and Non-Smokers

(Age wise)

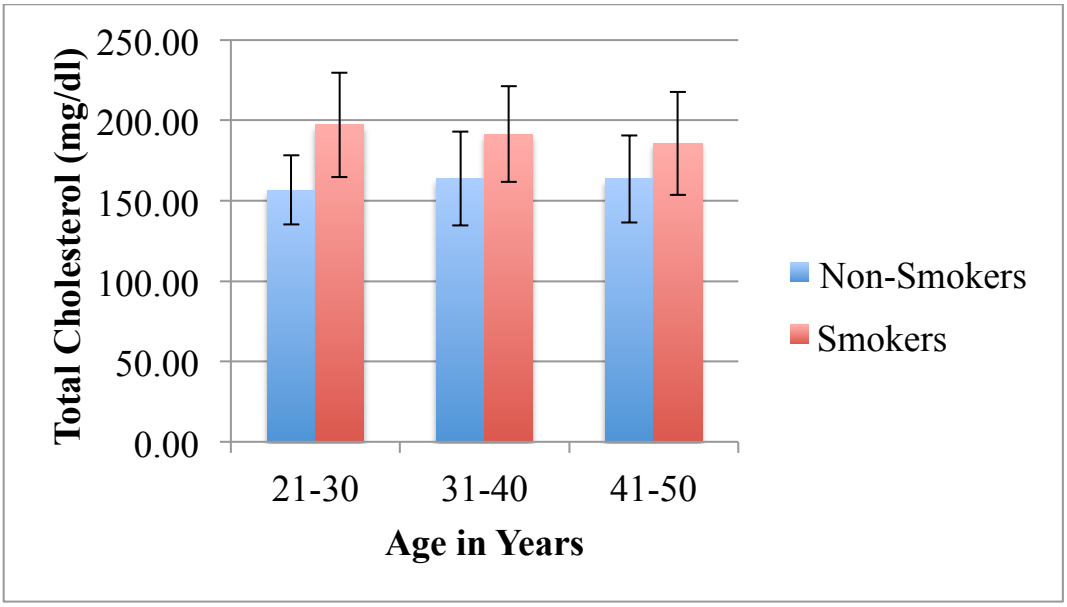


Table: 9

Comparison of TG among Non-Smokers and Smokers

(Age wise)

	Non-Smokers		Smokers	
Age in Years	No. of Persons	TG	No. of Persons	TG
21-30	36	101.75±21.51	30	171.33±29.16
31-40	43	103.65±29.53	49	163.08±25.88
41-50	21	106.57±26.3	21	157.05±32.97

Figure: 23

Comparison of TG among Non-Smokers and Smokers

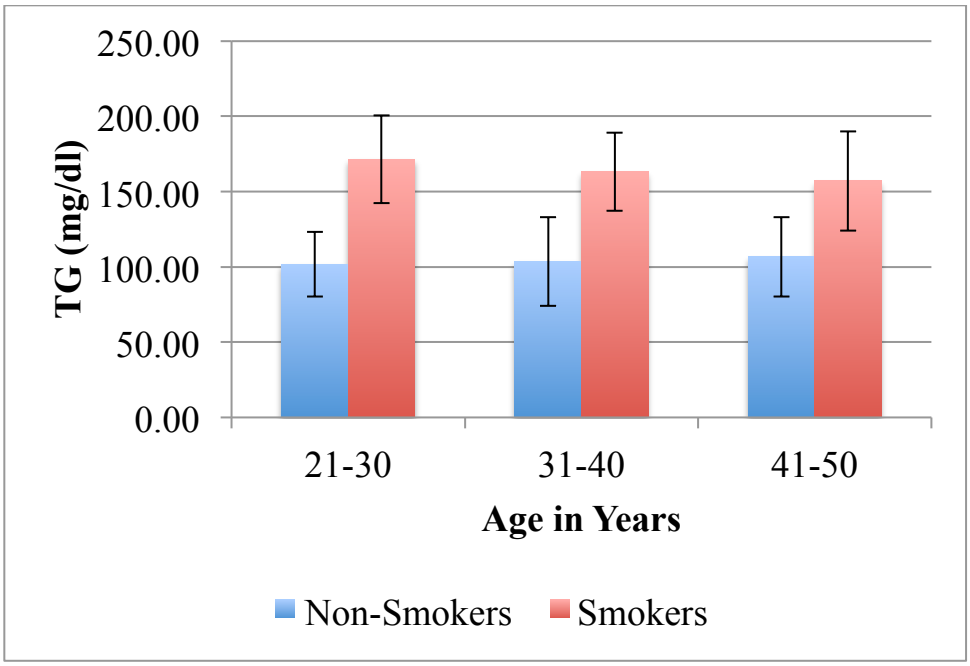


Table: 10

Comparison of HDL among Non-Smokers and Smokers

(Age wise)

	Non-Smokers		Smokers	
Age in Years	No. of Persons	HDL	No. of Persons	HDL
21-30	36	50.47±7.84	30	42.47±10.68
31-40	43	48.88±9.34	49	44.98±8.96
41-50	21	49.48±7.96	21	47.33±10.38

Figure: 24

Comparison of HDL among Non-Smokers and Smokers

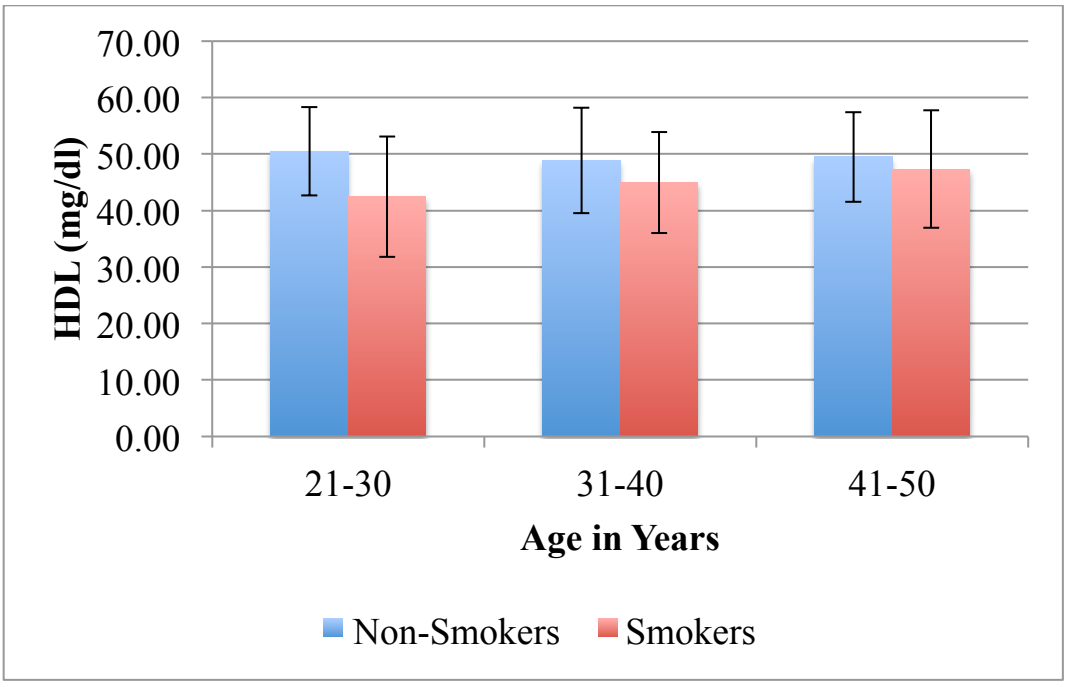


Table: 11

Comparison of LDL among Non-Smokers and Smokers

(Age wise)

	Non-Smokers		Smokers	
Age in Years	No. of Persons	LDL	No. of Persons	LDL
21-30	36	80.44±16.55	30	107.23±19.95
31-40	43	83.84±15.67	49	103.18±18.59
41-50	21	82.52±17.99	21	96.9±14.88

Figure: 25

Comparison of LDL among Non-Smokers and Smokers

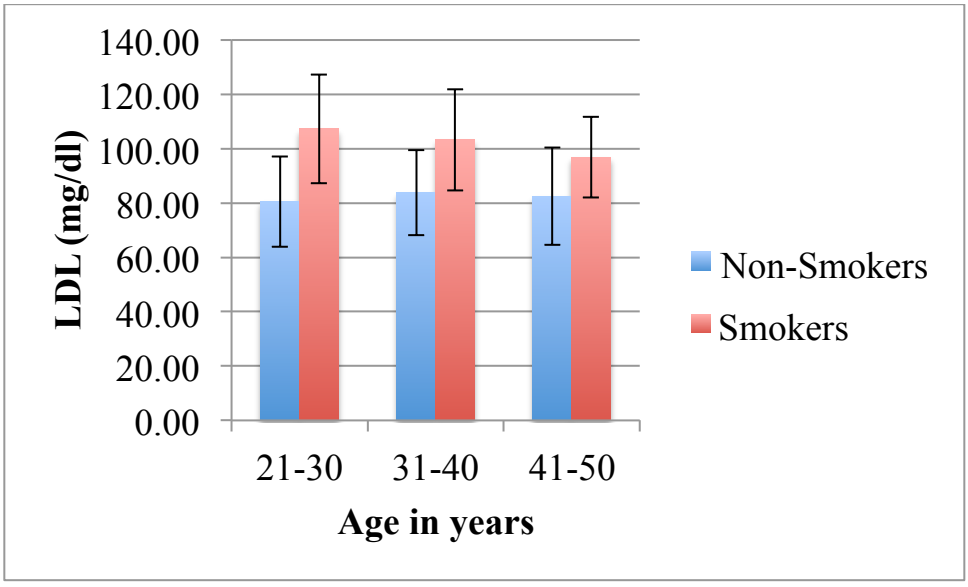


Table: 12

Comparison of VLDL among Non-Smokers and Smokers

(Age wise)

	Non-Smokers		Smokers	
Age in Years	No. of Persons	VLDL	No. of Persons	VLDL
21-30	36	20.44±6.2	30	30.7±10.12
31-40	43	23.21±6.39	49	28.73±8.56
41-50	21	20.71±6.21	21	27.29±7.73

Figure: 26

Comparison of VLDL among Non-Smokers and Smokers

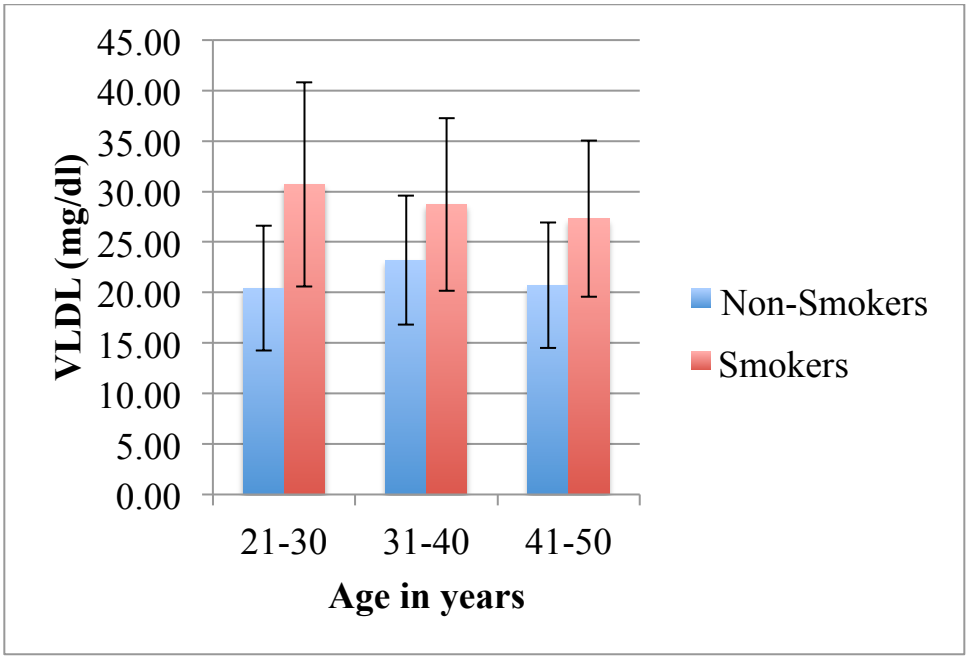


Table: 13

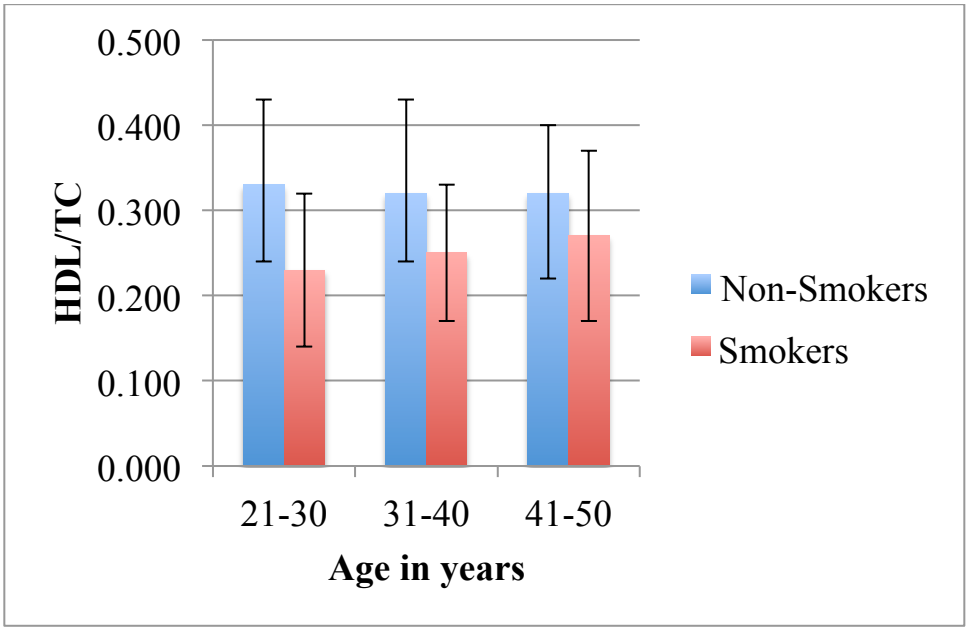
Comparison of HDL/TC among Non-Smokers and Smokers

(Age wise)

	Non-Smokers		Smokers	
Age in years	No. of Persons	HDL/TC	No. of Persons	HDL/TC
21-30	36	0.33±0.1	30	0.23±0.09
31-40	43	0.32±0.11	49	0.25±0.08
41-50	21	0.32±0.08	21	0.27±0.1

Figure: 27

Comparison of HDL/TC among Non-Smokers and Smokers



6. DISCUSSION

Cigarette smoking is found to occur mostly in later age group in Indian population when compared to western society. Third decade of life accounts for maximum number of smokers. In late adolescent periods and during college life they get captivated by the " Kick of smoke". Some start smoking to show off their macho image. Majority gets addicted to smoke but some people quit. Some go for other additive habits like cannabis. Smoking beedi is found mostly among the labor class in India. Smoking filtered tip cigarettes is found mostly among elite population.

Several studies have been conducted both in India and abroad among the smokers and they analyzed their clinical, biochemical and pathological parameters. The present study was conducted with the aims of comparing the lipid profile among smokers and non-smokers and to study changes in lipid profile depending on the intensity of smoking. This study was done as a part of postgraduate training programme.

Smokers are at a higher risk of CAD when compared to non-smokers. The various explanations given for this association includes alterations in blood coagulation, , reduced fibrinolysis, impaired arterial integrity and altered lipid profile and lipoprotein levels. The last explanation is examined in this study. Since in India smoking is less prevalent among females this

study was done only among male smokers. This discussion is planned to evaluate each variables separately.

Comparison between smokers and non-smokers of different age groups:

TOTAL CHOLESTEROL:

In the age group of 21 to 30 (30 persons) the mean total cholesterol value was 197.17 in smokers, while in non-smokers it was 156.64 (36 persons). In smokers of age group 31 to 40 (49 persons) the mean total cholesterol was 191.45 while in non-smokers it was 163.84(43 persons). The mean total cholesterol was 185.71 in smokers (21 persons) of age group 41 to 50 years while it was 163.52 in non-smokers (21 persons). The association of an increased total cholesterol value with smoking was noticed when smokers were compared with non-smokers of the similar age group. The mean total cholesterol value was 191.96 in smokers (100 persons) and 161.18 in non-smokers (100 persons), P value <0.001.

A study this kind comprising a very large number (that is more than 50,000 participants) was done in 1988 by the American Health Foundation of USA. Cholesterol screening was made across the country. The data collected from 10 screening centers were combined. The mean cholesterol values and standard errors were calculated for each both sexes and for various age groups. The results of this study are given below:

1. For each sex the mean cholesterol values increased with age.
2. For men, the mean cholesterol values increased gradually up to the age of 50 years, after which it appeared to be a plateau and then fell slightly. In both sexes mean cholesterol values decreased slightly after the age of 70 years, representing a possible survivorship effect.
3. It was noted that with an increase in the severity of smoking there was a statistically significant increase in mean total cholesterol values. Among men in age group of 20-60, they found 0.34 mg/dl increase in total cholesterol for every cigarette smoked. In women strongest association was observed in age interval between 30 to 50 years, where serum cholesterol increased by 0.5 mg/dl for each cigarette smoked.

These results indicated that heavy smokers of both sexes had considerably higher levels of plasma cholesterol than non-smokers, with the trend among women smokers aged 30-50 years greater than that for men and other women. For example, men under 50 years of age, who smoked 30 cigarettes or more per day had a predicted level approximately 10mg/ dl higher than those who never smoked; and women in the 30-50 age group had 15mg/dl values higher than non smokers.

The AHF study had certain limitations. The cholesterol screening was designed to be rapid: statistical analysis depended on the accuracy of self-reporting and the variables that might affect serum cholesterol levels were not collected. The explanation put forward by the AHF study were;

1. Repressive effects on oestrogen levels resulting in higher cholesterol level.
2. Differing pattern of inhalation in different age groups.
3. Consuming low fiber containing cereals and foods and dietary increase of fat, coffee and alcohol.
4. Sympathetic stimulation induced by nicotine leading to catecholamine release and further lipolysis.

The present study is in accordance with the AHF study, as a linear relationship is seen in the data collected; the total cholesterol levels and the severity of smoking. Karvonen et al also has noted such a correlation between smoking and elevation of cholesterol.

This study is also in accordance with study done by R.K. Tilwani, B. sapra, P.K.Sharma, R.K.Goyal in J.L.N.Medical college and A.G.Hospital, Ajmer, which showed increasing total cholesterol and severity of smoking. Similar results were noted by A.K.Sinha, G.C.Mishra, D.K.Paatel in their study on the same in V.S.S. Medical college, Orissa. Similar results were

observed in the study done by R.Rastogi and others in L.L.R.M. Medical College.

HDL cholesterol:

Of the smokers in 21-30 years of age group the mean value of HDL cholesterol stood 42.27mg/dl while the non-smokers showed a mean value of 50.47. Age related decrease was observed in the advancing age group probably due to increase in the duration of smoking. Smokers in the age group 31-40 years had mean HDL value of 44.98 mg/dl (49 persons), which is low compared to non-smokers of the same age group 48.88mg/dl (43 persons). In the age group of 41-50 years the smokers had a mean HDL value of 47.33mg/dl (21 persons) while the non-smokers of the same age group had a mean HDL value of 49.48 (21 persons). The mean HDL in smokers was 44.72 mg/dl (100 persons) while the mean of non smokers was 49.58 mg/dl (100 persons). P value =0.002.

Chemical pathology and medicine department of St. Marys hospital, London conducted a study and put forward an explanation to this effect. The action of endothelial lipoprotein lipase on triglyceride rich lipoprotein reduces the core volume of these particles and generates surface remnants containing unesterified cholesterol, phospolipid and apoprotein, which join the HDL3 pool. These surface remnants are thought to constitute the major

source of HDL precursors and lipoprotein lipase is thought to be a major determinant of plasma concentration of HDL. Esterification of the acquired free cholesterol in HDL3 particles by lecithin cholesterol acyltransferase results in the accumulation of cholesterol ester in the core of the particles and the production of larger, less dense HDL2 particles.

A transfer protein mediates the exchange of cholesterol ester in HDL with triglyceride in lipoprotein of a lower density. The magnitude of postprandial lipaemia determines the extent of this exchange and resultant triglyceride content of HDL2 particles. Furthermore, the triglyceride content of HDL2 particles has been shown to determine, which of the phospholipase or triglyceride lipase activities of hepatic lipase act on them. Triglyceride rich HDL2 particles are converted to HDL3 particles by removal of the triglyceride from the core of the particles, whereas phospholipid is removed from the surface of triglyceride poor HDL2 particles without any change in size or density.

Reduced intravascular lipolysis exists in smokers, hence it was suggested that a consequent increase in the postprandial lipaemia in smokers will result in a greater proportion being converted to HDL3 particles by hepatic lipase. This would explain the lowering of HDL2 and the increase in the concentration of HDL3 cholesterol found in smokers.

HDL2 concentration correlates negatively with the severity of angiographically defined atheroma. The change in HDL that is reported in young smokers and the increased exposure of vascular endothelium to the potentially atherogenic lipoproteins as a consequence of impaired clearance of triglyceride rich lipoproteins is the mechanism whereby smoking predisposes to coronary artery disease.

The present study had not focused on HDL values following cessation of smoking. But data published by Moffatt R.J. et al and Quensel et al revealed normalization of HDL in smokers who gave up that habit. HDL values increased by 7mg/dl in 48days after stopping smoking and the effect disappeared in people who resumed the habit.

The present results are in concordance with R.K. Tilwani et al study in J.L.N. Medical college and A.G. Hospital, Ajmer and also with study done by R.Rastogi and others in L.L.R.M. Medical college. A.K. Sinha and others reported similar results in their study conducted in V.S.S. Medical college, Orissa.

LDL-cholesterol:

Quantitative estimation of LDL in this study showed a mean value of 107.23mg/dl in smokers of 21-30 years age group which comprised 30 participants while the non smokers (36 persons) showed a value of

80.44mg/dl. The mean LDL values of smokers in the age group 31-40 (49 persons) was 103.18mg/dl and that of non smokers of the same age group (43 persons) was 83.84mg/dl. The mean LDL values of smokers of the age group 41-50 was 96.9mg/dl and that of non smokers was 82.52mg/dl. The LDL values were high in the smokers compared with non-smokers of the same age group. The mean LDL value in smokers was 103.08mg/dl when compared with non smokers which was 82.34mg/dl. P-value (<0.001).

To establish a causal relationship between exposure to cigarette smoke and changes in serum lipid and lipoprotein concentrations, dose-response effects were calculated by an analysis of 54 published studies conducted by wendy Y.F. and his associates from the foundation of Blood research, scarbrough, USA. They found a higher mean concentration of cholesterol (3%), triglycerides (9.1%), VLDL (10.4%), LDL (1.7%). The dose response effect on VLDL and LDL among non smokers, mild, moderate and severe smokers were (0, 7.2 , 44.4, 39.0%) and (0 , -1.1, 1.4 , and 11.0%) respectively.

This study results are comparable with LDL levels of R.Rastogi and other's study on the same topic. R. Rastogi et al study showed higher levels of LDL in heavy smokers. Similar results were reported by A.K. Sinha and

associates in their study of effect of cigarette smoking on lipid profile in the young.

TRIGLYCERIDES:

In the age group of 21-30 years smokers (30 persons) showed a mean value of 171.33 mg/dl while the mean value of triglyceride in non-smokers was 101.75mg/dl in the same age group. In the 31-40 years age group mean value among smokers was 163.08mg/dl (49 persons): non-smokers showed 103.65mg/dl (43 persons). The mean triglyceride value was 157.05mg/dl in smokers of age group 41-50 years (21 persons) and in non smokers it was 106.57mg/dl (21 persons) . The mean value of TG in smokers was 164.29mg/dl when compared to non-smokers, which was 103.58mg/dl. (p value <0.001). The triglycerides show a steady increase with increase in the severity of smoking.

The Craig WY study on persons in 8-19 age groups had shown an increase of 11.8% in smokers for the triglyceride value while their analysis of the 54 published data showed an increase in triglyceride value of 9.1% in smokers compared to non-smokers.

Sympatho adrenal system stimulation by nicotine leads to lipolysis and increased serum free fatty acid levels which lead to increased synthesis of VLDL from the liver and hence triglycerides.

These findings are also similar to those done by R.K. Tilwani (J.L.N. Medical college) and R. Rastogi L.L.R.M. Medical college. A.K. Sinha et al showed significant increase of LDL cholesterol in severe smokers.

HDL to Total cholesterol:

Several studies which have evaluated the risk factor for ischaemic heart disease has showed that reduced HDL–cholesterol to total cholesterol is a very important risk factor. The present study showed significant reduction in the HDL-C/TC ratio in smokers compared to non-smokers. The values in non smokers of age group 21-30, 31-40, 41-50 were 0.33±0.1, 0.32±0.11, and 0.32±0.08, While in smokers of the same age group the values were 0.23±0.09, 0.25±0.08, 0.27±0.1 respectively which was significantly low. This is also in accordance with studies conducted abroad and in India. A Comparative study of HDL-c/TC ratio showed in 0.25 in smokers when compared to 0.32 in non-smokers.

VLDL:

The VLDL values of smokers of the age group 21-30, 31-40,41-50 years were 30.7, 28.73, 27.29 mg/dl respectively. While the values in non-smokers of the similar age group were 20.44, 23.21, 20.71mg/dl respectively. The mean value in nonsmokers was 21.69 mg/dl while that of

smokers was 29.02 mg/dl . P value (<0.001). VLDL values show a steady increase with the severity of smoking.

Comparision of values depending on the severity of smoking:

The mean total cholesterol in mild smokers was 183.06±33.38 while that of moderate smokers was 194.09±29.39, and that of severe smokers was 209±29.15. The cholesterol values were found to increase with increase in the severity of smoking. The mean triglyceride value of mild smokers was 155.06±31.80, while that of moderate and severe smokers were 166±26.63 and 176.5±25.62 respectively. With the increase in the severity of smoking there is increase in the triglyceride values. The mean HDL values of mild, moderate and severe smokers were 48.03±8.65, 45.06±10.26 and 42±9.67 respectively. The HDL values show a steady decline with increase in the severity of smoking. The mean LDL values in mild, moderate and severe smokers were 97.12±13.05, 105.58±20.07, and 114±20.41 respectively. The values of LDL show a steady increase with increase in severity of smoking. The mean VLDL of mild, moderate and severe smokers were 23.15±7.67, 32.73±6.67 and 34±9.27 respectively which show an increase in values with increased severity of smoking.

The mean total cholesterol value of non-smokers was 161.18±26.77 while that of smokers was 191.96±31.37. The mean triglyceride value of

non-smokers was 103.58±26.26 while in smokers it was 164.29±28.95. The mean HDL value of non-smokers was 49.58±8.57 while in smokers it was 44.72±9.96. The mean LDL values of non-smokers was 82.34±16.57 while in smokers it was 103.08±18.66. The mean VLDL value of non-smokers was 21.69±6.42 while in smokers it was 29.02±8.98. The total cholesterol, triglyceride, LDL and VLDL values were significantly elevated in smokers when compared to the non-smokers. The HDL values were significantly lower in smokers when compared to the non-smokers. When HDL-C/TC ratio of smokers were 0.25 compared with that of non smokers which was 0.32 , significantly lower levels were found in moderate and severe smokers.

7. CONCLUSION

- Increase in total cholesterol, triglycerides, low-density lipoproteins and very low-density lipoproteins were found in smokers of all age group, whereas high-density lipoproteins showed an inverse relationship.
- A direct relationship exists between the severity of smoking and increase in the total cholesterol, triglycerides, LDL and VLDL while an inverse relationship is found with HDL.
- Asymptomatic smokers are at the risk of developing coronary artery disease due to the above changes in lipid profile.
- The observed values were in concordance with the studies done in India and other countries.

8. LIMITATIONS OF THE STUDY

Since the population selected for this study was small and the study is not a blinded study, an element of observer bias may have occurred. Large population studies should be conducted to confirm the results.

9. SUMMARY

The present study was carried out among 200 patients who attended Madurai Rajaji Government Hospital. 100 smokers and 100 non-smokers were selected for this study. The study was conducted from July 2014 to September 2014.

- All the patients studied were males.
- The population was selected in the age group of 20 to 50 years.
- Among non-smokers the mean value of total cholesterol was 161.18±26.77, triglycerides was 103.58±26.26, HDL was 49.58±8.57, LDL was 82.34±16.57 and VLDL was 21.69±6.42.
- Among mild smokers the mean value of total cholesterol was 183.06±33.38, triglycerides was 155.06±31.80, HDL was 48.03±8.65, LDL was 97.12±13.05 and VLDL was 23.15±7.67.
- Among moderate smokers the mean value of total cholesterol was 194.09±29.39, triglyceride was166±26.63, HDL was 45.06±10.26, LDL was 105.58±20.07 and VLDL was 32.73±6.67.

- Among severe smokers the mean value of total cholesterol was 209±29.15, triglycerides was 176.50±25.62, HDL was42±9.67, LDL was 114±20.41 and VLDL was 34±9.27.

BIBLIOGRAPHY

1. Ezzati M, Henley SJ, Thun MJ, Lopez AD. Role of smoking in global and regional cardiovascular mortality (Circulation 2005). Circulation 2005; 112:489.
2. Paul M. Ridker and Peter Libby. Risk factors for atherothrombotic disease. In: Braunwald's heart disease – A textbook of cardiovascular medicine. 8th Edn, New Delhi: Published by Elsevier, 2008. p.1004.
3. David M. Burns Nictoine addiction chapter 375, Harrison's principles of internal medicine, Eds, Dennis L. Kasper, Eugene Braunwald, Anthony S. Fauci, Stephen L. Hauser, Dan L. Lango, J Larry Jameson, 16th Edn., Vol.2, McGraw Hill, 2005 .p.2573-76.
4. Alexi A. Wright and Ingrid T. Katz. Tobaco tightrope-balancing disease prevention and economic development in China. N-Eng J Med 2007 Apr; 356: 1493-1496.
5. WHO. WHO/WPRO Smoking statistics. 28' May 2002.
6. www.wpro.who.int/media-centre/fact-sheets/fs-2002.
7. WHO. WHO report on the global tobacco epidemic 2008. http: WWW:WHO.int/tobacco/m_power/mpower_report_prevalence_data.2008.pdf.

8. Jindal SK, et al. Tobaco smoking in India: prevalence quit rates and respiratory morbidity. Indian Chest Dis Allied Sci 2006; 48: 37-42.

9. WHO. The tobacco atlas. www.who.int/entity/tobaco/en/atlas4.pdf.

10. US Department of Health and Human Services. 11th Report on carcinogens by the US Department of Health & Human Services. http://ntp:niehs.nih.gov/ntp/roc/tocc//.html

11. US Department of Health and Human Services centers for disease control and prevention: National center for chronic disease prevention and Health promotion office on smoking and health. 2004; 134: 824-826.

12. Satyanarayana U. Text book of biochemistry reprinted. June 2001: 29-42.

13. Executive summary of the third report of NCEP expert panel on detection evaluation and treatment of blood cholesterol in adults. Adults Treatment Panal 3rd JAMA 2001; 285:2486.

14. Sidharth Das Vmohan. Disorders of lipid metabolism API textbook of medicine. Ed. Siddharth N. Shah. API 2003; 250-256.

15. Robert K. Murray, Daryl K. Branner, Victor W. Rodwell. Harper's illustrated biochemistry. 27th Edn., Kothleem M. Botham. 2003. p.217-40.

16. Rader DJ, Hobbs HH. Disorders of lipoprotein metabolism – Harrison's principles of internal medicine. 17th Edn., Vol.II, .p.2416-2428.
17. Sing G. The effect of smoking on serum lipids. JAPI 2002 Dec; 50: 1494.
18. Lipids on line slide library. www.lipidsonline.org
19. Abbassi Abdulah et al. Effect of chronic cigarette smoking on lipid profile. Pak J Med Health Sci 2007 Mar; 1(1): 35-6.
20. National cholesterol education program. Executive Summary of the third report of the national cholesterol education. Program expert Panel on Detection, Evaluation and Treatment of High Blood Cholesterol in Adults JAMA 2001; 285:2486-2497.
21. K. Park. Principles of epidemiology and epidemiological methods. Chpater-3, Parks text book of preventive and solid medicine. 18th Edn, M/s Banarasidas Bhanot Publsihers 2005. p.73-74.
22. NS Neki. Lipid profile in chronic smokers – A clinical study. Journal of Indian Academy of Clinical Medicine. 2002 Jan-Mar; 3:51-54.
23. Heitzer J, Meinertz T. Prevention of coronary artery disease. Smoking Pub Med 2005; 94(3:III): 30-42.

24. Guedas DP, Guedes JE, Barbosa DS, de Oliveria JA. Tobaco use and plasma lipid – lipoprotein profile in adolescents. Pub Med 2007 Jan-Feb; 53: 5963.

25. Ioannis N, Mammas, George K. Bertsias, Manalis linardakis, Nikolaos E. Tzamkis, Demetre N. Labadanios. Cigarette smoking: alcohol consumption, and serum lipid profile among medical students in Greece the European. Journal of Public Health 2003; 13(3):278-782.

26. Pauk A, Lopes, Maria C, Santhoas. Livin vicante and Ana M Viegs – Crespo. Clirica Chimica Acta. 2004; 348(1-2): 49-55.

27. Kharb S, Singh GP. Effect of smoking on lipid profile lipid peroxidation and antioxidant status in normal subjects and in patients during and after acute myocardial infarction. Clin Chim Acta 2000 Dec; 302: 213-9.

28. Khurana M< Sharma D, Khamdelwal PD. Lipid profile in smokers and tobacco chewers – A comparative study. J Assoc Physicians India 2000 Sept; 48(9): 895-7.

29. Libby Peter. The vascular biology of athrosclerosis. Braunwald's heart disease. 7th Edn, 2005; 35: 921.

30. Folsom AR, et al. Determinants of population changes in fibrinogen and factor VII over 6 years. The atherosclerosis risk in communities (ARIO) study: Atherosclerosis, thrombosis, and vascular biology. 2000; 20(2): 601-6.
31. Willigondael EM et al. Influence of smoking on incidence and prevalence of peripheral arterial disease. J Vasc Surg 2004 Dec; 40(6): 1158-65.
32. Creager MA. The relationship of cigarette smoking to peripheral arterial disease. Rev Cardiovasc Med 2004; 5(4): 189-93.
33. Jiang Y, et al. Prevalence of peripheral arterial disease and its association with smoking in a population based study in Beijing, China. J Vascul Surg 2002; 44(2): 333-338.
34. Das UN, Fames EFA. The brain lipid heart connection interaction between exercise, insulin, cytokines, free radicals, and nitric oxide and its relevance to coronary artery disease. The Association of Physicians of India Medicine Update 2002 ed. SB Gupta, 12: 217-219.
35. Newby DE. Impaired coronary tissue plasminogen activator. Release is associated with coronary atherosclerosis and cigarette smoking circulation. 2001; 103(15): 1936-41.

36. Mantosh Panja. Will aggressive lipid lowering replace coronary intervention the association of physicians of India. Medicine Update 2002;12: 243-245.

37. Bolego C, Poli A, Paoletti R. Smoking and gender cardiovascular Res. 2002 Feb; 53; 568-76.

38. Ambrose JA, Barua RS. The pathophysiology of cigarette smoking and cardiovascular disease. J Am College of Cardiology 2004 May; 431:1731-17.

39. Wenzel S, Schorr K, Degenhardt H, Frischkopf K, Kojda G, Wiesner RJ. Rosenkranz S, Piper HM, Schluter K-D, TG-B, down regulates PTHrp in coronary endothelial cells. J Mol Cell Cardiol 2001; 33:181-90.

40. Kasap, S., A. Gonenc, D.E. Sener and I. Hisar. Serum cardiac markers in patients with acute myocardial infarction: oxidative stress, C-reactive protein and N-terminal probrain natriuretic peptide. J. Clin. Biochem. Nutr., 2007;41: 50-57.

41. Anazawa, T., PC Dimayuga, H. Li, S. Tani, J. Bradfield, K.Y. Chyu, S. Kaul, P.K. Shah and B. Cercek. Effect of exposure to cigarette smoke on carotid artery intimal thickening: the role of inducible NO synthase. Arterioscler. Thromb. Vase. Biol., 2004; 24: 1652-1658.

42. Fagerstrom, K. The epidemiology of smoking: health consequences and benefits of cessation. Drugs, 2002; 2: 1-9.

43. Panagiotakos, D.B., L.S. Rallidis, C. Pitsavos, C. Stefanadis and D. Kremastinos. Cigarette smoking and myocardial infarction in young men and women: a case-control study. Int. J. Cardio., 2007;116: 371-375.

44. Zornoff, L.A.M., D.R. Duarte, M.F. Minicucci, P.S. Azevedo, B.B. Matsubara, L.S. Matsubara, A.O. Campana and S.A.R. Paiva. Effects of beta-carotene and smoking on heart remodeling after myocardial infarction. Arq. Bras. Cardiol., 2007;89:151-157.

45. Orhan, H., C.T.A. Evelo and O. Sahin. Erythrocyte antioxidant defense response against cigarette smoking in humans - the gutathione S-transferase vulnerability. J. Biochem. Mol Toxicol., 2005;19: 226-233.

46. Muzakova, V., R. Kandar, P. Vojtisek, J. Skalicky, R. Vankova, A. Cegan and Z. Cervinkova. Antioxidant vitamin levels and glutathione peroxidase activity during ischemia/reperfusion in myocardial infarction. Physiol. Res., 2001; 50: 389-396.

47. Kharb, S., G.P. Singh. Effect of smoking on lipid profile, lipid peroxidation and antioxidant status in normal subjects and in patients during and after acute myocardial infarction. Clin Chim Acta, 2000; 302: 213-219.

48. Waart, F.G., T.J. Smilde, H. Wollersheim, A.F.H. Stalenhoef and F.J. Kok. Smoking characteristics, antioxidant vitamins, and carotid artery wall thickness among life-long smokers. J. Clin. Epidemiol., 2000;53: 707-714.

49. Anbarasi, K., G. Vani, K. Balakrishna, Shyamala Devi, C.S. Effect of bacoside a on brain antioxidant status in cigarette smoke exposed rats. Life. Sci., 2006; 78: 1378-1384.

50. Yamaguchi, Y., S. Kagota, J. Haginaka and M. Kunitomo. Evidence of modified LDL in the plasma of hypercholesterolemic WHHL rabbits injected with aqueous extracts of cigarette smoke. Environ. Toxicol. Pharmacol., 2000;8: 255-260.

51. Ambrose, J.A. and R.S. Barua. The pathophysiology of cigarette smoking and cardiovascular disease. J. Am. Coll. Cardiol., 2004; 43: 1731-1737.

52. NS Neiki. Lipid profile in chronic smokers – A clinical study. JIACM 2002;3(1):51-54.

53. Jose A. De Velasco, Juam A. Ratrigurz Francisco Ridocci, Jose Aznar. Action to improve secondary prevention in coronary heart disease patients. European Heart Journal Supplements 2004; 6: 227-229.

54. Libby P. Current concepts of the pathogenesis of the acute coronary syndrome. Circulation 2001; 104:365-379.

55. Beerian K, Kis Z, Virok D, et al. Independent and joint effects of antibodies to human heart shock protein 60 and Chlamydia pneumonia infection in the development of coronary atherosclerosis. Circulation 2001; 103:1503-1508.

56. Rai JL, Demaster EG, Jaimes EA. Cigarette smoke induced endothelium dysfunction. Role of superoxide anion. J Hypertens 2001; 19:891-897.

57. Jantunen R, Juvonen E, Ikkala E, Oksanen K, Anttila P, Ruutv T. The predictive value of vascular risk factors and gender for the development of thrombotic complications in essential thrombocytemia. Ann Hematol 2001; 80:74-78.

58. Tietz NH. Clinical guide to laboratory tests. 4th Edn., Philadelphia : W.B. Saunders Company; 2001 .p.268-269.

59. Cole TG. Measurement of triglycerides concentration - handbook of lipoprotein testing. Washington DC; AACC Press; 2000. p.115-126.

60. Assmann G. Lipid diagnostic Hente. In: Greten H. et al., Lipoprotein and herzinfarkt Witzstrock – Verlag, Baden – Baden /W. Germany. 2001 .p.29.

61. Tilwani, Mjos DO, Norway T. Lipid effects of smoking. Am Heart J 2003; 115:272-275.

62. Sinha AK, Misra GC, Patil DK. Effects of cigarette smoking on lipid profile in the young. JAPI 2002; 43:185-188.

63. Rastogi R, Srivastava S, Mehrotra, Gupta MK. Lipid profile in smokers. JAPI 2001;37(12):764-767.

64. Tiwari AK, Gode JD, Dubey GP. Effect of cigarette smoking on serum total cholesterol and high-density lipoprotein cholesterol in normal subjects and coronary heart patients. Indian Heart J 2001;41(2):92-94.

PROFORMA

A COMPARATIVE STUDY OF LIPID PROFILE AMONG SMOKERS AND NON-SMOKERS

Case No : OP No :

Name :

Age / Sex :

Occupation :

Religion :

Address :

Height :

Weight :

Date of Sampling :

1. **PRESENTING COMPLAINS:**
2. **HISTORY OF PRESENTING ILLNESS:**

(Type, onset, duration, progression, present state of the complaints, associated features if any)

3. **PAST HISTORY:**

HT / DM / PTB / BA /I.H.D /Renal disease / Liver disease / Hyperlipidemia /Thyroid disorders /other

4. FAMILY HISTORY:

- Hyperlipidemia
- Death in the family due to diseases related to smoking
- Death in the family due to ischemic heart disease

5. DRUG HISTORY:

If any specify, drug _____ dose _____ duration _____

6. PERSONAL HISTORY

Sleep :

Appetite :

Diet : Vegetarian/Non-vegetarian

Preference for fatty food in routine diet : Yes/No

Bowel : Bladder :

History of smoking : Yes/No

Cigarettes/day :

Beedies/day :

Years of smoking : (duration)

History of alcohol in take : Yes/No since

History of betel nut chewing : Yes/No since

History of tobacco chewing : Yes/No since

History of any other habits : Yes/No since (If Yes Specify)

7. GENERAL EXAMINATION:

a) Built : Well/moderately/poorly

b) Nourishment : Under nourished/Normal/obese

c) Anemic[pallor] : Yes/No

d) Jaundice[icterus] : Yes/No

e) Clubbing : Yes/No

f) Oedema : Yes/No

g) Lymphadenopathy : Yes/No

h) Eyes: Arcus Senilis : Yes/No

i) Thyroid : Normal/Enlarged

j) Opticfundus : Normal /
Hypertensive /Diabetic

k) BP :

l) Pulse : Rate :
Rhythm :

Character :

Volume :

Condition of the vessel wall :

m) J.V.P :

n) Respiratory rate :

o) Temperature :

p) Body mass index :

8. SYSTEMIC EXAMINATIONS:

a) Cardiovascular examination:

Inspection:

- Shape of the chest /Any deformity
- Precordium
- Apical impulse
- Visible pulsation

Palpation:

- Apex beat : Size
  Character
  Thrill
  Palpable sounds
- Left parasternal heave : Present/absent
- 2nd left intercostal space
  Palpable s2 : Thrill :
- Other areas :

Auscultation

S1,S2 : Normal/Abnormal/ Specify

S3 : Yes/No

S4 : Yes/No

Murmurs : Absent/Present
Specify

b) Respiratory system : Normal/Abnormal

If abnormal specify

c) Gastro-intestinal system : Normal/Abnormal
If abnormal specify

d) Nervous system : Normal/Abnormal

If abnormal specify

e) Genitourinary system : Normal/Abnormal

If abnormal specify

9.INVESTIGATIONS:

a) Fasting Blood Sugar :

b) Post Prandial Blood Sugar :

c) Blood urea :

d) Serum creatinine :

e) Lipid profile

- Total Cholesterol:
- LDL :
- HDL :
- Serum Triglycerides :
- VLDL :

f) Thyroid Profile :

g) Electrocardiogram :

MASTER CHART

NON-SMOKERS

S.NO	Name	AGE (yrs)	TC	TG	HDL	LDL	VLDL
1	Akbar	25	161	111	57	80	24
2	Aandy	40	137	101	55	79	30
3	suresh	30	123	92	51	78	19
4	Mohamed Ali	37	180	159	38	108	28
5	Ajith	29	102	81	56	63	12
6	Arumugam	32	145	100	58	85	25
7	Chinna Mayan	26	143	118	46	90	23
8	Eswaran	36	167	122	42	99	31
9	Ganesh	45	187	163	36	126	28
10	Hariharan	23	201	170	40	151	40
11	Ram Chandar	34	128	72	56	70	22
12	Jeya Singh	38	163	130	49	85	27
13	Nagaraj	49	150	102	47	90	16
14	Irulandi	23	125	75	49	90	22
15	Dharmaraj	39	175	87	53	112	28
16	Kesavan	30	144	130	46	88	20
17	Chandra Mouli	32	192	166	40	109	31
18	Bhagiyaraj	34	121	69	53	75	14
19	Alagar Samy	23	166	120	48	99	28
20	Alaudeen	31	161	90	49	85	20
21	Karuppa Samy	24	176	85	43	92	28
22	Kulanthai Velu	40	155	78	50	67	19
23	Maharajan	43	133	66	54	54	14
24	Mohan Kumar	32	188	123	36	85	25
25	Rajkumar	23	166	103	49	76	18
26	Sivakumar	32	197	106	38	99	29
27	Ramar	45	154	87	43	61	17
28	Ravi	37	258	169	30	115	40
29	Aswin	25	177	98	47	78	29
30	Singamuthu	39	182	101	34	94	24
31	Essakki	26	153	84	54	89	18
32	Veera Kumar	36	199	109	33	103	28
33	Savior	44	142	79	49	71	16

S.NO	Name	AGE (yrs)	TC	TG	HDL	LDL	VLDL
34	Chandhana Mariappan	21	172	98	45	86	22
35	Santhanam	40	165	95	50	83	21
36	Alagu	36	137	60	61	54	16
37	Ponnu samy	26	149	76	56	60	19
38	Prabhakar	48	151	89	50	74	20
39	Rathinam	32	189	109	42	94	23
40	Mayandi	42	162	101	48	88	21
41	Ruthreshwar	27	169	94	53	81	17
42	Manikkam	25	141	85	51	80	12
43	Priyadharsan	23	170	116	32	92	19
44	Maniratham	46	240	172	30	112	30
45	Panneer Selvam	39	162	89	47	91	27
46	Rahuman	25	148	82	58	63	12
47	Rajesh	40	139	76	59	59	15
48	Surya	21	160	99	39	75	19
49	Senthil Ganesh	32	157	90	42	62	20
50	Manikandan	28	170	117	45	69	13
51	Subramani	33	129	70	61	67	19
52	Arunachalam	49	183	127	51	83	25
53	Daniel Rajan	22	165	97	42	79	17
54	Selvam	29	180	128	44	93	26
55	Kannan	50	172	98	53	90	28
56	Prasanna	35	156	99	48	76	16
57	Marimuthu	33	149	96	52	79	19
58	Kumar	30	170	111	58	87	24
59	Veerachamy	50	203	138	36	107	37
60	Muthu Veeran	33	168	95	49	75	26
61	Irrulandi	47	130	81	53	58	15
62	Maha Kumar	25	183	118	30	92	29
63	Janardhanan	23	120	71	60	55	14
64	Alphonse	39	194	142	33	96	31
65	Antony Raj	41	177	103	52	85	20
66	Ahamed Mohideen	32	160	100	50	89	18

S.NO	Name	AGE (yrs)	TC	TG	HDL	LDL	VLDL
67	Moorthi	30	186	131	48	91	31
68	Sivanantha prabhu	27	155	93	59	88	20
69	Manivannan	37	183	129	40	92	28
70	Sathyamoorthi	27	136	87	58	70	14
71	Sankaranarayana	31	190	144	46	96	29
72	Raja	28	158	96	52	71	16
73	Rakesh	43	168	90	44	81	21
74	Murugan	33	122	58	63	60	17
75	Dharmaprabhu	39	197	135	42	93	27
76	Indrian	28	169	89	52	62	26
77	Veeriyan	38	144	92	49	82	18
78	Gnaprakash	46	195	128	49	99	23
79	Mookandi	33	120	90	64	66	13
80	Janakarajan	25	141	110	62	71	15
81	Biju	44	173	104	54	97	26
82	Boominathan	32	130	98	59	68	14
83	Gowthan	37	185	106	53	80	23
84	Samy	49	151	101	58	74	18
85	Samuthirakani	29	176	120	59	88	22
86	Anbarasu	38	140	81	60	65	24
87	Deva	32	153	89	60	70	20
88	Thiagarajan	24	178	84	52	63	16
89	Soundara Pandi	50	145	76	61	75	18
90	Selvakumar	32	122	56	62	68	15
91	Bhootham	27	165	114	56	69	18
92	Velu	39	171	144	52	80	20
93	Alagu Raja	43	132	111	51	64	11
94	Ragupathi	22	140	124	56	77	21
95	Michel	45	128	110	59	68	16
96	Rajendran	40	170	90	52	95	20
97	Joesph	39	140	74	58	81	18
98	Narasiman	21	101	56	64	60	13
99	Vijay	43	158	112	61	76	15
100	Akilesh	31	225	168	34	114	40

SMOKERS

MILD SMOKERS

S.NO	Name	AGE (yrs)	TC	TG	HDL	LDL	VLDL
1	Rajkumar	34	170	151	50	92	20
2	Maha Krishna	41	254	221	31	119	31
3	Mahesh	50	150	130	53	88	15
4	Veerachamy	33	138	115	55	89	21
5	Usman	39	220	197	32	120	32
6	Pitcahai	44	188	158	54	100	20
7	Rangan	22	127	103	59	76	19
8	Selva Alagar	40	175	138	57	94	21
9	Singaraj	45	199	172	46	97	38
10	Palanivel	36	220	196	32	103	32
11	Muthalagan	30	171	139	49	100	16
12	Karthikeyan	33	182	149	55	99	21
13	Krishnanmoorthy	23	196	164	48	99	20
14	Rajasekar	37	176	145	54	89	19
15	Athidya	34	184	156	42	90	21
16	Vikraman	47	195	160	46	93	18
17	Udayakumar	22	168	149	57	96	23
18	Prabhu	31	292	251	31	144	45
19	Devaraj	36	162	143	49	92	16
20	Srinivasan	29	183	147	42	94	21
21	Ravikumar	33	143	146	56	98	18
22	Sekar	49	190	164	52	97	21
23	Abubacker	25	149	143	60	85	21
24	Balaji	36	186	151	47	94	17
25	Justin	29	208	190	34	108	32
26	Thangapandi	39	192	158	51	103	19
27	Sridhar	46	116	65	61	75	14
28	Manimaran	35	180	153	49	98	22
29	Ramu	30	167	142	50	96	18
30	Babu Ganesh	40	179	146	49	87	32
31	Vivekanandan	34	212	167	34	116	42
32	Asan Mohideen	21	189	151	52	94	19
33	Sudhakar	32	180	157	48	80	20

MODERATE SMOKERS

S.NO	Name	AGE (yrs)	TC	TG	HDL	LDL	VLDL
34	Solomon	29	192	148	51	120	29
35	Mohamed Faizal	35	198	162	45	132	26
36	Muthu Vel	27	182	165	44	101	23
37	Ravi Varman	21	201	159	38	122	40
38	Mohan Prasanth	34	156	132	52	78	26
39	Nagarajan	44	188	145	54	92	32
40	PetchiMuthu	40	234	192	31	129	31
41	Vinodan	23	140	132	59	85	27
42	Mari Selvan	31	170	149	52	78	31
43	Siva Raman	26	233	198	34	131	45
44	Anoop Chandran	32	182	152	49	111	37
45	Prakash	30	205	188	33	107	38
46	Senthoor Pandi	34	222	194	31	134	46
47	Alagesan	29	193	171	54	120	37
48	Tamil Kumaran	45	178	146	49	97	26
49	Rajarathan	35	228	182	36	127	43
50	Sankaralingan	49	187	151	58	97	33
51	Rajasingh	39	179	149	55	88	26
52	Praveen	24	242	196	28	122	32
53	Sambantham	39	190	161	46	102	26
54	Alhaj	33	189	167	52	92	32
55	Velraj	29	243	204	29	125	39
56	Udayam	34	270	248	28	146	48
57	Kalimuthu	40	167	146	41	89	29
58	Amudan	47	209	199	35	100	36
59	Oliyarasu	41	159	146	53	95	25
60	Arjun	31	189	158	55	115	32
61	Pankaj Kumar	27	192	170	39	98	38
62	Manoj	37	134	112	60	68	23
63	Devadoss	22	181	160	57	62	27
64	Mannan	32	170	150	51	96	28
65	Ramasamy	27	213	198	32	124	34
66	Mathivaanan	37	189	148	56	101	35

SEVERE SMOKERS

S.NO	Name	AGE (yrs)	TC	TG	HDL	LDL	VLDL
67	Manivanna Bhoopathi	38	230	196	30	130	36
68	Raja Sekaran	28	178	150	55	82	22
69	Jeya Chandram	40	200	176	35	112	30
70	Arumuga Nainar	29	180	179	39	92	23
71	Vishnu Chandar	31	198	165	34	99	28
72	Nagendran	45	256	217	26	143	43
73	David	27	172	148	43	94	19
74	Paraman	33	145	143	52	80	14
75	Bakthavatsalam	29	256	221	28	141	41
76	Anbumani	34	181	160	50	93	17
77	Thilagaraj	50	169	151	56	88	20
78	Vasudevan	28	213	190	34	118	31
79	Mokkasamy	39	197	159	39	98	27
80	Gopinath	30	244	219	30	132	49
81	Sellakannu	45	169	149	57	91	24
82	Thirumalai	40	196	162	45	87	33
83	Vinayakam	30	243	199	33	139	50
84	Ravi	35	199	179	45	129	41
85	Kirubakaran	44	190	160	35	86	35
86	Lakhmi Narayanan	50	225	199	32	123	31
87	Mathusudhanan	38	185	160	48	92	19
88	Vikaram kumar	29	203	189	34	112	46
89	Kathiravan	35	177	159	49	89	29
90	Chinnadurai	50	150	145	59	78	23
91	Siddarth	40	193	156	44	95	36
92	Sundar	30	251	228	29	142	42
93	Thiruvengadam	32	202	181	32	125	29
94	Manokaran	42	174	145	51	89	25
95	Linkeshwaran	36	187	150	47	101	22
96	Peter	47	160	121	52	88	27
97	Sathesh	37	239	196	31	140	40
98	Arokiaraj	50	194	154	34	99	36
99	Ramesh	33	206	171	38	114	38
100	Mahadevan	37	188	157	54	98	32

Ref.No.6506/E1/5/2014

Madurai Medical College,
Maduai -20. Dated: 11.09.2014.

Institutional Review Board/Independent Ethics Committee
Capt.Dr.B.Santhakumar,MD (FM). deanmdu@gmail.com
Dean, Madurai Medical College &
Government Rajaji Hospital, Madurai 625 020 . Convenor

Sub: Establishment – Madurai Medical College, Madurai-20 –
Ethics Committee Meeting – Meeting Minutes - for August 2014 –
Approved list – reg.

The Ethics Committee meeting of the Madurai Medical College, Madurai was held on 05th August 2014 at 10.00 Am to 12.00 Noon at Anaesthesia Seminar Hall at Govt. Rajaji Hospital, Madurai . The following members of the Ethics Committee have attended the meeting.

1.Dr.V.Nagarajan,M.D.,D.M(Neuro) Ph: 0452-2629629 Cell No.9843052029 nag9999@gmail.com.	Professor of Neurology (Retired) D.No.72, Vakkil New Street, Simmakkal, Madurai -1	Chairman
2.Dr.Mohan Prasad, MS.M.Ch. Cell.No.9843050822 (Oncology) drbkcmp@gmail.com	Professor & H.O.D of Surgical Oncology (Retired) D.No.32, West Avani Moola Street, Madurai.-1	Member Secretary
3. Dr.L.Santhanalakshmi, MD (Physiology) Cell No.9842593412 dr.l.santhanalakshmi@gmail.com.	Vice Principal, Prof. & H.O.D. Institute of Physiology Madurai Medical College	Member
4.Dr.K.Parameswari, MD(Pharmacology) Cell No.9994026056 drparameswari@yahoo.com.	Director of Pharmacology Madurai Medical College.	Member
5.Dr.S.Vadivel Murugan, MD., (Gen.Medicine) Cell No.9566543048 svadivelmurugan_2007@rediffmail.com.	Professor & H.O.D of Medicine Madurai Medical College	Member
6.Dr.A.Sankaramahalingam, MS., (Gen. Surgery) Cell.No.9443367312 chandrahospitalmdu@gmail.com	Professor & H.O.D. Surgery Madurai Medical College.	Member
7.Mrs.Mercy Immaculate Rubalatha, M.A., Med., Cell.No.9367792650 lathadevadoss86@gmail.com	50/5, Corporation Officer's Quarters, Gandhi Museum Road, Thamukam, Madurai-20.	Member
8.Thiru.Pala.Ramasamy, B.A.,B.L., Cell.No.9842165127 palaramasamy2011@gmail.com	Advocate, D.No.72,Palam Station Road, Sellur, Madurai-20.	Member
9.Thiru.P.K.M.Chelliah, B.A., Cell No.9894349599 pkmandco@gmail.com	Businessman, 21 Jawahar Street, Gandhi Nagar, Madurai-20.	Member

The following Project was approved by the Ethical Committee

Name of P.G.	Course	Name of the Project	Remarks
Dr.Naseema Banu.S.H. naseeshahul@gmail.com	PG in MD (General Medicine) Madurai Medical College & Rajaji Hospital, Madurai	**A comparative study of lipid profile among smokers and non smokers.**	**Approved**

Please note that the investigator should adhere the following: She/He should get a detailed informed consent from the patients/participants and maintain it Confidentially.

1. She/He should carry out the work without detrimental to regular activities as well as without extra expenditure to the institution or to Government.
2. She/He should inform the institution Ethical Committee, in case of any change of study procedure, site and investigation or guide.
3. She/He should not deviate the area of the work for which applied for Ethical clearance. She/He should inform the IEC immediately, in case of any adverse events or Serious adverse reactions.
4. She/He should abide to the rules and regulations of the institution.
5. She/He should complete the work within the specific period and if any Extension of time is required He/She should apply for permission again and do the work.
6. She/He should submit the summary of the work to the Ethical Committee on Completion of the work.
7. She/He should not claim any funds from the institution while doing the work or on completion.
8. She/He should understand that the members of IEC have the right to monitor the work with prior intimation.

Member Secretary
Ethical Committee

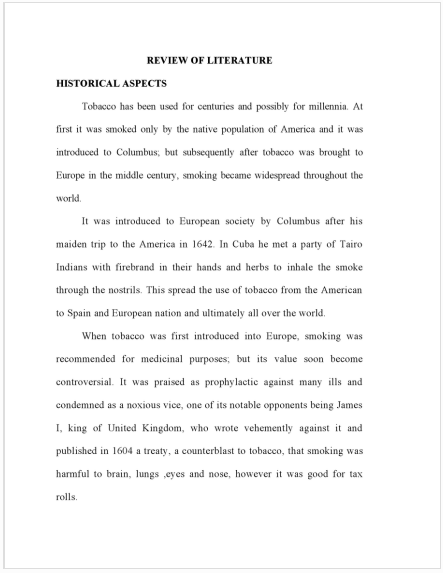
Chairman
Ethical Committee

11.9.14

DEAN/Convenor
Madurai Medical College & Govt.
Rajaji Hospital, Madurai- 20.

27.6
11/9/14

To
The above Applicant
-thro. Head of the Department concerned



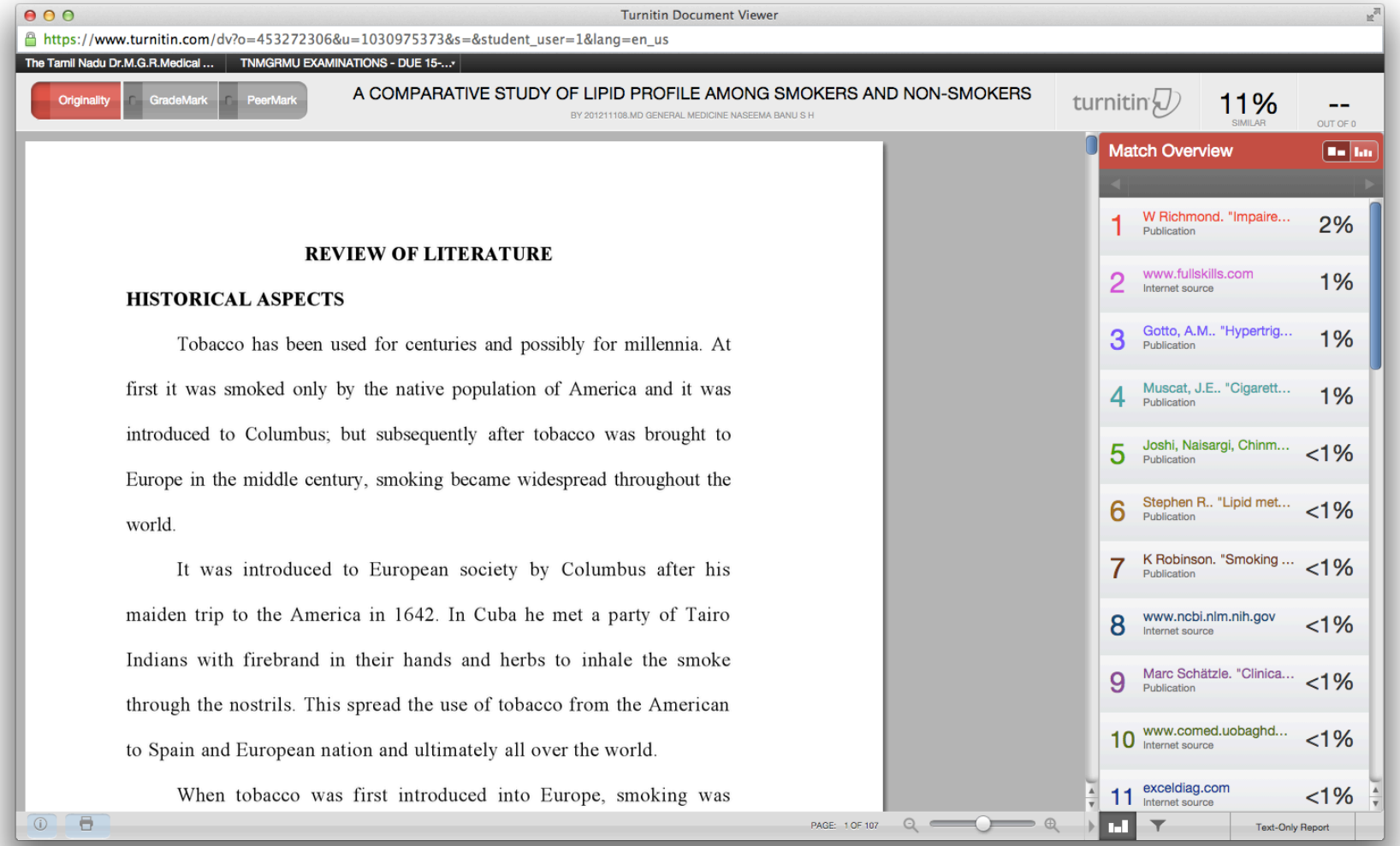
REVIEW OF LITERATURE

HISTORICAL ASPECTS

Tobacco has been used for centuries and possibly for millennia. At first it was smoked only by the native population of America and it was introduced to Columbus; but subsequently after tobacco was brought to Europe in the middle century, smoking became widespread throughout the world.

It was introduced to European society by Columbus after his maiden trip to the America in 1642. In Cuba he met a party of Tairo Indians with firebrand in their hands and herbs to inhale the smoke through the nostrils. This spread the use of tobacco from the American to Spain and European nation and ultimately all over the world.

When tobacco was first introduced into Europe, smoking was recommended for medicinal purposes; but its value soon become controversial. It was praised as prophylactic against many ills and condemned as a noxious vice, one of its notable opponents being James I, king of United Kingdom, who wrote vehemently against it and published in 1604 a treaty, a counterblast to tobacco, that smoking was harmful to brain, lungs ,eyes and nose, however it was good for tax rolls.


Turnitin Document Viewer
https://www.turnitin.com/dv?o=453272306&u=1030975373&s=&student_user=1&lang=en_us
The Tamil Nadu Dr.M.G.R.Medical ...
TNMGRMU EXAMINATIONS - DUE 15-...
Originality
GradeMark
PeerMark
A COMPARATIVE STUDY OF LIPID PROFILE AMONG SMOKERS AND NON-SMOKERS
BY 201211108.MD GENERAL MEDICINE NASEEMA BANU S H
turnitin
11%
SIMILAR
--
OUT OF 0
REVIEW OF LITERATURE
HISTORICAL ASPECTS
Tobacco has been used for centuries and possibly for millennia. At first it was smoked only by the native population of America and it was introduced to Columbus; but subsequently after tobacco was brought to Europe in the middle century, smoking became widespread throughout the world.
It was introduced to European society by Columbus after his maiden trip to the America in 1642. In Cuba he met a party of Tairo Indians with firebrand in their hands and herbs to inhale the smoke through the nostrils. This spread the use of tobacco from the American to Spain and European nation and ultimately all over the world.
When tobacco was first introduced into Europe, smoking was
PAGE: 1 OF 107
Match Overview
1 W Richmond. "Impaire... Publication 2%
2 www.fullskills.com Internet source 1%
3 Gotto, A.M.. "Hypertrig... Publication 1%
4 Muscat, J.E.. "Cigarett... Publication 1%
5 Joshi, Naisargi, Chinm... Publication <1%
6 Stephen R.. "Lipid met... Publication <1%
7 K Robinson. "Smoking ... Publication <1%
8 www.ncbi.nlm.nih.gov Internet source <1%
9 Marc Schätzle. "Clinica... Publication <1%
10 www.comed.uobaghd... Internet source <1%
11 exceldiag.com Internet source <1%
Text-Only Report

Turnitin Originality Report

A COMPARATIVE STUDY OF LIPID PROFILE AMONG SMOKERS AND NON-SMOKERS by 201211108.md General Medicine NASEEMA BANU S H

From TNMGRMU EXAMINATIONS (The Tamil Nadu Dr.M.G.R.Medical Uty 2014-15 Examinations)

Processed on 27-Sep-2014 21:10 IST
ID: 453272306
Word Count: 15197

Similarity Index	Similarity by Source	
11%	Internet Sources:	4%
	Publications:	8%
	Student Papers:	3%

sources:

1 2% match (publications)
W Richmond, "Impaired intravascular lipolysis with changes in concentrations of high density lipoprotein subclasses in young smokers.", BMJ, 07/25/1987

2 1% match (Internet from 09-Apr-2009)
http://www.fullskills.com/smoking/stats.php

3 1% match (publications)
Gotto, A.M., "Hypertriglyceridemia: Risks and perspectives", The American Journal of Cardiology, 19921214

4 1% match (publications)
Muscat, J.E., "Cigarette smoking and plasma cholesterol", American Heart Journal, 199101

5 < 1% match (publications)
Joshi, Naisargi, Chinmay Shah, HB Mehta, and PA Gokhle, "Comparative study of lipid profiel on healthy smoker and non smokers", International Journal of Medical Science and Public Health, 2013.

6 < 1% match (publications)
Stephen R., "Lipid metabolism and secondary forms of dyslipoproteinemia in children", Progress in Pediatric cardiology, 200309

7 < 1% match (Internet from 22-May-2013)
http://www.ncbi.nlm.nih.gov/pubmed/16482950

8 < 1% match (publications)
K Robinson, "Smoking and acute coronary heart disease: a comparative study.", Heart, 12/1/1988

9 < 1% match (publications)
Marc Schätzle, "Clinical course of chronic periodontitis: effect of lifelong light smoking (20 years) on loss of attachment and teeth : Light smoking and attachment loss", Journal of Investigative and Clinical Dentistry, 07/14/2010

10 < 1% match (Internet from 21-May-2014)
http://www.comed.uobaghdad.edu.iq/uploads/Lectures/Medicine%204/Atherosclerotic%20heart%20disease%20&%20peripheral%2

11 < 1% match (Internet from 06-Oct-2013)
http://exceldiag.com/catalogs/11034.pdf

12 < 1% match (publications)
S. F. KNUTSEN, "Wives of coronary high-risk men - are they also at higher risk? The Tromsø Heart Study", Journal of Internal Medicine, 10/1990

13 < 1% match (publications)
Mjos, O.D., "Lipid effects of smoking", American Heart Journal, 198801